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NORTH CAROLINA

AGRICULTURAL EXPERIMENT STATION

OF THE

COLLEGE OF AGRICULTURE AND MECHANIC ARTS

WEST RALEIGH

A SERIOUS LETTUCE DISEASE AND A METHOD OF CONTROL

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N. C. COLLEGE OF AGRICULTURE AND MECHANIC ARTS

THE NORTH CAROLINA

AGRICULTURAL EXPERIMENT STATION

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N. C. AGRICULTURAL EXPERIMENT STATION,
WEST RALEIGH, N. C.

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A SERIOUS LETTUCE DISEASE (SCLEROTINIOSE) AND A METHOD OF CONTROL*

By F. L. Stevens and J. G. Hall.†

PART I.

Lettuce, formerly a garden plant grown for home consumption, has of recent years become an important article of farm production, representing, says Mr. L. F. Kinney,[1] "a higher form of agriculture than any known to previous generations." Its production has so increased that in 1897 we find the statement that the "sales of this vegetable from a single farm during the last fifteen years have amounted to over a half million dollars."

This crop has been grown commercially in the United States for more than fifty years, and was the first crop to be grown extensively under glass by market gardeners.[1]

As early as 1872, it was estimated that no less than 50,000 sashes were used mainly for this purpose within ten miles of Boston.[2]

About 800 acres are now devoted to seed production in California alone, making some 400,000 pounds of seed.‡

Lettuce was first grown largely on a commercial scale in Connecticut and Rhode Island. It was estimated that in 1893 fully nine-tenths of the winter head lettuce sold in New York and other eastern markets was either grown in Rhode Island or in the vicinity of Boston.[3]

During the last decade there has been a large increase in the shipment from the South to the metropolitan markets, particularly from North Carolina and South Carolina where lettuce is grown under cloth, and from Florida where it is grown largely in the open. In Florida it was first cultivated under canvas about 1894, Mr. F. D. Warner, of Gainesville, and Mr. Denby being among the pioneers in this industry in that State, as was also Mr. J. E. Pace, of Sanford, who introduced the crop in that place in 1896.

Lettuce is now grown to a large extent in South Carolina, particularly at Conway where it was first grown about the year 1900. Charleston and James Island are other prominent lettuce centers in South Carolina.

The Lettuce Industry in North Carolina.

Wilmington was the pioneer lettuce growing community of North Carolina, and the first lettuce raised under cloth for shipment in this State seems to have been grown by D. W. Trask, of Wilmington, about 1892, three years before other commercial lettuce was produced in Wilmington. Mr. Trask, who raised lettuce in a small way for the

*The matter here published has in part been presented at various scientific meetings, American Phytopathological Society, Boston, 1909; the N. C. Academy of Science, Greensboro, May, 1908; the N. C. Academy of Science, Raleigh, May, 1911, and in part published in Bulletin No. 217, N. C. Agr. Exp. Sta.

†Mr. G. W. Wilson has taken the place of Mr. Hall during the last year of the work.

‡Letter from Mr. W. W. Gilbert, Bureau Plant Industry, who gives his information as coming from W. W. Tracy, Sr., of the Horticultural Office.

home market, had more than could be sold there and was forced to ship it. This lettuce sold at from $6.00 to $10.00 a barrel and the area under cloth was increased the next year to more than an acre. After the third year others began raising this profitable crop, and the area gradually increased until it now aggregates between 75 and 100 acres under cloth.

The leading lettuce growers of the Wilmington section are: W. H. Mills & Sons, F. D. Klein, W. E. Springer, H. L. Thorne, D. W. Trask, O. Martindale, C. E. Kerr, A. O. McEarchern, B. B. Trask, Moses Horne, F. T. Kerr, C. F. Seitter, Add. Hewlett, and Geo. W. Trask.

Around New Bern lettuce was first grown for market in 1894, by Mr. W. H. Bray with an area of about three acres, and Messrs. Hackburn and Willet with one acre.

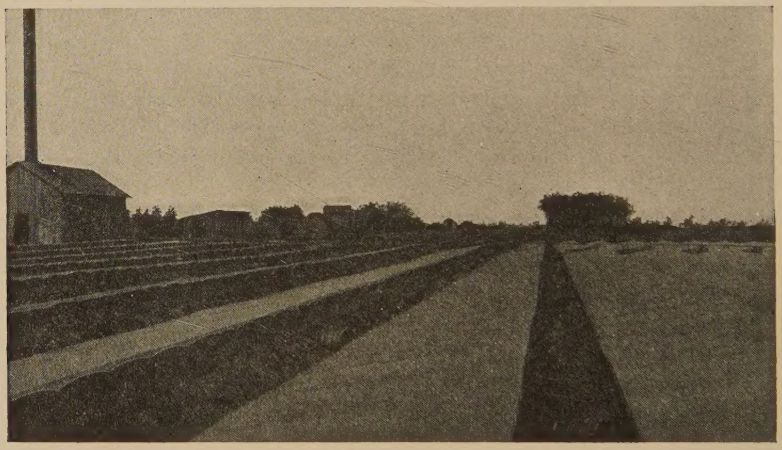


FIG. 1.—Lettuce as grown under canvas with irrigation at New Bern, N. C.

The acreage in the immediate vicinity of New Bern, all under cloth and irrigated and much of it steam heated, soon increased manyfold.

The most extensive growers of lettuce around New Bern are or have been, Hackburn & Willet, W. H. Bray, Edward Clark, J. M. Spencer, Thomas Daniels, H. H. Tooker, and J. A. Meadows.

At Fayetteville, commercial lettuce culture was apparently begun about 1895 by Fittzell Brothers with two thousand plants and the industry has increased rapidly, the crop at times totalling between 15 and 50 acres.

The principal growers are, or have been, W. H. Tomlinson, J. A. Pemberton, S. H. Strange, H. T. Drake, J. A. Nicholson, William Kyle, W. L. Hawley, Fittzell & Fittzell and many small producers, more than forty-five in all.

At Warsaw, about 1897, L. Middleton and J. A. Powell raised lettuce commercially. H. H. Caroton, Will Corbett, O. P. Middleton, J. A. Powell, Sr., J. A. Powell, Jr., Henry Middleton, of Warsaw;

and John Hamilton, Charlie Gore, Rob Wells, and Charlie Pickett, of Magnolia, are, or have been, prominent in the industry.

At Maxton, lettuce was grown for shipment in 1902 by H. C. McNair and H. S. McNair.

Considerable lettuce is also grown for shipment at Faison, Willard, Wade, Tarboro, Chadbourn, and Mt. Olive.

Characteristic Symptoms of Sclerotiniose.

Sclerotiniose may readily be distinguished from any other lettuce disease when the specific symptoms are once known.

Of these symptoms the one which first catches the eye of the lettuce grower is the rotting of his plant in whole or in part. When first observed a single leaf may be drooping, or wilting; a day or so later the whole plant appears involved, the outer leaves dropping flat on

Fig. 2.—Plant showing drop, one symptom of sclerotiniose.

the ground, the central head alone remaining standing. At this stage the plant appears as though scalded by an application of hot water. The head also soon succumbs to the rot and topples over. The first conspicuous symptom is this rotting and "dropping" of the whole plant.

Close examination of such rotting plants, especially in the later stages of the disease, reveals the presence of a delicate weft of cotton-

like threads on the underside of the affected leaves, especially in the more moist regions as at the base of the leaves near the stem. This character is limited to Sclerotiniose, and is a sure indication of this disease.

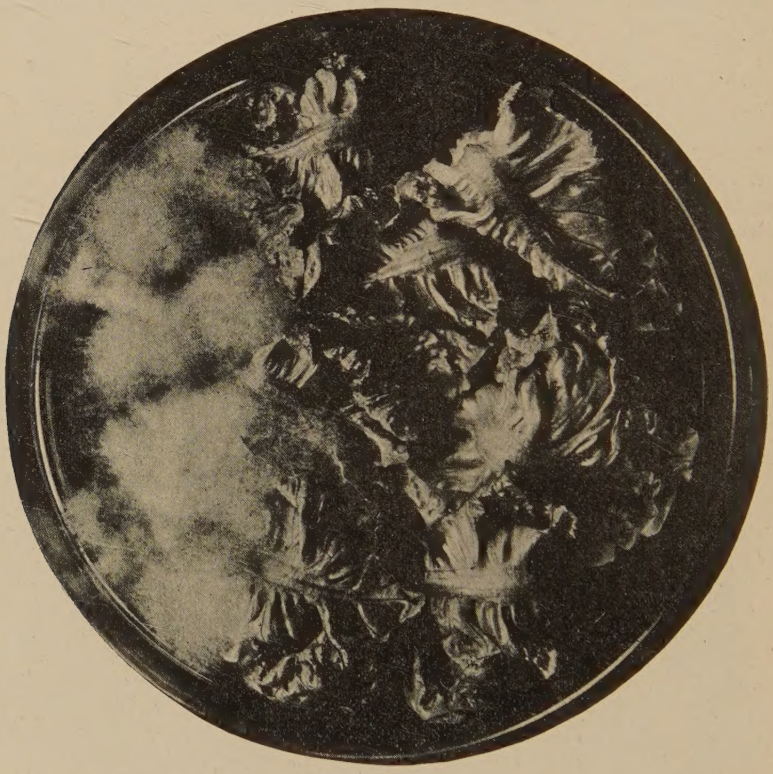


FIG. 3.—Sclerotiniose: mycelium growing upon leaves in culture dish. This cotton-like weft of mycelium is definitely characteristic.

In the last stages of the disease, a week or two after the final dropping of the plant, there will be found many small black bodies, varying in size from that of a pin head to a grain of corn, in, or upon, or under the sick portions of the plant. These too are absolutely characteristic of Sclerotiniose.

These three characteristics—the dropping, the cotton-like mycelium, and the sclerotia—if carefully observed, enable anyone to pronounce with certainty as to whether or not a given bed or plant is affected with Sclerotiniose.

HISTORY OF SCLEROTINIOSE IN AMERICA.

The disease characterized by the symptoms indicated above is termed *sclerotiniose* from the fungus *Sclerotinia* which is its cause.

Since one of the chief symptoms of Sclerotiniose, the symptom which certainly first catches the eye of practical lettuce growers, is a dropping

and rotting of the outer leaves, followed usually by dropping and rotting of the rest of the plant, this disease has come to be called "the drop," by lettuce growers in many sections of the country. These symptoms may be produced by several different causes.[4]

"The drop" is therefore not one single definite disease. It is rather a condition or a symptom just as lameness of horses is a condition or a symptom, not a disease. Lameness may be due to spavin, which is one disease, or to tuberculosis, which is another, etc. So the drop may be due to *Sclerotinia,* to *Pythium,* to *Botrytis,* etc., each of these causing a separate disease and each requiring different treatment and prophylaxis according to the nature of its cause.

The first definite knowledge of the existence of lettuce *Sclerotiniose* is contained in a communication by Smith in 1900.[4]

The disease is there first clearly and accurately characterized and attributed to its causal fungus, *Sclerotinia libertiana* Fckl. While 1900 is thus the earliest date of accurate knowledge concerning this disease, it was in all probability seriously injurious long before that time, and many serious lettuce troubles reported from different parts of the United States, and attributed to other causes, were doubtless really due to Sclerotinia. Sclerotiniose frequently occurs in conjunction with other lettuce diseases and in many instances inroads upon the lettuce beds attributed to *Botrytis, Rhizoctonia, Bacteria,* or other causes, were probably due in part, even in main, to sclerotiniose, often doubtless complicated by one or more of these other diseases. Among such early, but somewhat uncertain cases are the following:

Prof. G. E. Stone, of Massachusetts, says in a letter to the senior author (April, 1908), "I think we have evidence to show that the disease has been here a number of years, and that it is not a native in this region. It was doing considerable damage in the lettuce houses in the 90's—I think the fungus is not indigenous as it does not occur in some of our greenhouses in this State. I know many greenhouses which became infected with the drop through the introduction of plants from the Boston market-garden district. When I first studied the disease I had to introduce it into my greenhouse, and I knew of a number of houses at that time which never had it.

"I do not believe the disease troubled the very early lettuce growers, and I imagine it was not severe in the 90's. I do not believe the men who grew lettuce for 40 or 50 years had much trouble in growing it under sash or even in their old greenhouses. My predecessor in the station, Prof. Humphrey, I think probably had the same trouble under observation when he was here in 1888 or 1889, and probably some of the other observers who described the bacterial disease of lettuce, had the drop. I have always considered that Sclerotinia and eel worms were both introduced organisms, and were not indigenous to our State. What I have said in regard to the sclerotium being absent from some houses for many years, also applies to eel worms, since there are many houses which have never been troubled with these. I look upon many of these diseases as simply the result of extensive commercial relations with foreign countries."

J. E. Humphrey[5] in 1892 records a lettuce disease in Massachusetts which Smith[4] thought "covers what is now generally known in the lettuce district as 'the drop,'" though there is no real certainty that Humphrey had actually to do with sclerotiniose. The only evidence that his disease was such is that it occurs in a region where this disease was subsequently very prevalent, and that his description agrees with that of the drop.

L. H. Bailey[6] in 1895 pictured a lettuce plant which in the picture appears to be a typical case of drop. He attributed it to *Botrytis,* not to *Sclerotinia,* though it is possible that *Sclerotinia* was present and remained unnoticed.

A. D. Selby[7] in 1896 mentioned a disease as "lettuce rot" attributing it to *Botrytis.* This may have been a form of drop, and was possibly due in part of *Sclerotinia,* though there is no evidence that anything but *Botrytis* was present.

G. E. Stone and R. E. Smith[8] in 1897 described a disease which they called "the drop" and which they attributed to *Botrytis.* Though from their later paper it seems possible that this early outbreak was in part at least due to *Sclerotinia.*

In 1898 Stone and Smith[9] and again in 1899[10] refer to an outbreak of lettuce "drop," still attributing it to *Botrytis,* though it probably was in part, even largely, due to *Sclerotinia.*

H. Garman in 1899[11] speaks of lettuce rot, which from his description, seems to have been some form of drop. *Botrytis* and other organisms were seen, but *Sclerotinia* was not definitely mentioned.

True *sclerotiniose* was mentioned and figured in Hume in 1901.[12] He then said "within the last few seasons a disease has wrought considerable destruction to the crop. This disease is commonly known among the growers as 'damp off.' In some cases the attack resulted in the total loss of the crop, while in others a loss of from 25 to 50 per cent. was suffered."

The disease is said by Rolfs in a letter to one of the authors to have been severe in the region of Gainesville about 1896, at which time a number of fields were almost completely destroyed.

Ramsey in 1904[13] described a lettuce disease due to the presence of *Botrytis* on the fall crop and a genuine case of *sclerotiniose* on the second crop in later winter. It was clearly characterized by the presence of cottony mycelium and by sclerotia.

In North Carolina, the drop, probably in all cases true sclerotiniose, first attracted the attention of lettuce growers around New Bern in 1897; at Fayetteville in 1901; at Warsaw in 1902. It was mentioned by the senior author of this bulletin in his Annual Report of 1907,[14] and was the subject of a press bulletin in the same year.[15]

It is briefly referred to by Hutt in a bulletin of the North Carolina Department of Agriculture.[16]

Sclerotiniose is now known to occur, as is shown in the accompanying map, in all of the South Atlantic States, North Carolina, South Carolina, Alabama, Georgia, and Florida, and also in Maine, Vermont, Massachusetts, New York, Connecticut, Rhode Island, Pennsylvania,

Delaware, Louisiana, Wisconsin, Washington, and Iowa, and a partially verified record of its occurrence in Virginia exists. While not definitely recorded from other States, it probably occurs in many of them, particularly near the Atlantic seaboard.

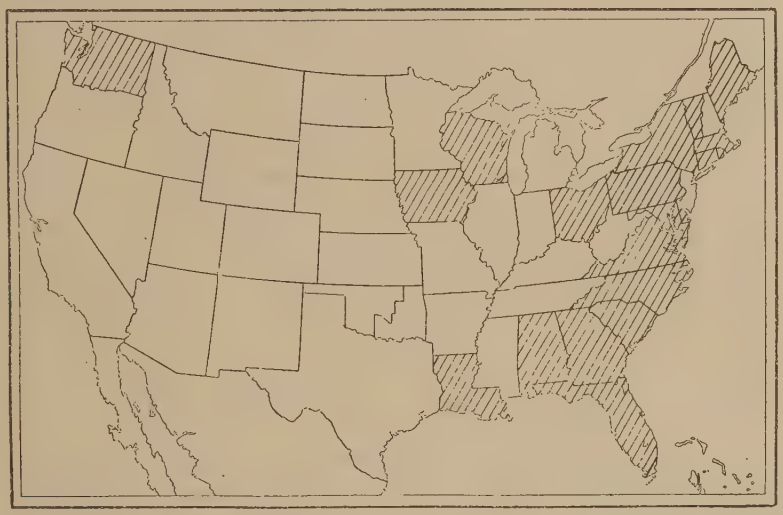


FIG. 4.—Map showing distribution of sclerotiniose in the United States. The disease is definitely reported in shaded States, but is not known to occur in others.

From what is known of the disease it is certain that it persists long and multiplies in territory once infected, and rapidly invades new regions. It is therefore increasing the area under its tribute yearly, and will continue to become of wider significance, especially as the lettuce industry broadens.

Summarizing the history of Scleriotiniose it may be said to have attracted attention first about 1890 in Massachusetts, in 1896 in Florida, in 1897 in North Carolina, in 1904 in Wisconsin, and to now possess practically the whole Atlantic seaboard and some of the more western States.

AMOUNT OF DAMAGE.

The extent of the damage varies with the severity of the epidemic and with the value of the crop affected. In 1900 in Massachusetts Stone and Smith[17] placed the proportion of plants succumbing to the disease at 15 to 85 or 95 per cent. of the crop. "The latter percentages are very exceptional, as growers are not content to experience this loss more than once without making radical changes in their methods. Practically entire crops have been destroyed by drop alone to our knowledge, and the majority of growers in Massachusetts have experienced at one time or another a loss of from 15 to 40 per cent. The loss of 25 per cent. from drop is no uncommon experience in a large number of lettuce houses, and when we consider that these houses each may contain from 6,000 to 12,000 plants, worth from 40 cents to $1.00 per dozen, some idea of the loss may be obtained."

In Florida* the loss is very severe and is sometimes complete. In South Carolina while crops are frequently destroyed and the lettuce industry seriously threatened as is shown in the following quotation from a personal letter.† "Several years ago I grew lettuce quite extensively for Northern markets, but had to give it up on account of the damping off. * * * Lettuce is not grown here as extensively as in former years principally on account of this disease."

In Maryland sclerotiniose does damage in many greenhouses.‡

In Alabama sclerotiniose is reported by Wilcox§ to do much damage in those places where lettuce is grown on a large scale. In Auburn and Montgomery it is repeatedly met, with great loss.

In New York, some growers, says Stewart** have had considerable trouble with the drop. It certainly is one of the troublesome diseases of lettuce." Whetzel, of the same State, says "it (the drop) occurs more or less commonly in all greenhouses about this State and sometimes in lettuce fields."

Amount of Damage in North Carolina.

In North Carolina the lettuce "drop" or "damp off" now shows itself to greater or less extent at New Bern, Wilmington, Maxton, Fayetteville, Willard, Raleigh, and probably at numerous localities where lettuce is of less importance. The damage done by it in 1906 is variously estimated at 10 per cent., 20 per cent., 33 per cent., 50 per cent., and 70 per cent. by different growers. At Fayetteville the damage from this disease is estimated at from 10 to 50 per cent. of the total value of the crop. Around Wilmington the loss is placed at 10 per cent. At New Bern estimates vary from 33 1-3 per cent. to 50 per cent., while at Maxton the loss is placed at 20 per cent.

The disease sometimes appears the first season the crop is grown in a given soil, often not until many crops have been raised. When once it does gain a foothold in a bed it persists, multiplies and increases until usually the grower is forced to move the bed to new regions, usually to very soon meet again a similar fate. Thus the loss to the crop is coupled with the loss attendant upon moving the lettuce bed, frames, irrigating, and heating pipes to new land and the leaving of the enriched soil to go to a newer and poorer one.

Cursory Description of Sclerotiniose.

The cause of the "drop" is a fungus belonging to the genus *Sclerotinia*, a genus which is well known on account of its many destructive species, among them being two that are particularly conspicuous, one causing a serious and widespread apple rot and the other causing one of the worst of peach diseases. The fungus is known technically as *Sclerotinia libertiana Fuckel.* It was first described in 1869, and bears its present name, libertiana, in honor of Marie Anne Libert

*Letter from P. H. Rolfs, March 30, 1908.
†D. T.West, Charleston, S. C., May, 1907.
‡Personal letter from J. B. S. Norton, December 14, 1906.
§Personal letter from E. M. Wilcox, March 28, 1908.
**F. C. Stewart in personal letter, March 30, 1908.

who published on parasitic fungi from 1813 to 1837. The plant body of this fungus consists of delicate branching, mold-like threads called the mycelium, which may, with the microscope or if abundant with the naked eye, be seen in or on the affected parts of the lettuce plant. No diseased part is free from them and they are, on the other hand, never present without being accompanied by a condition of disease in the adjacent parts of the lettuce plant. It has been definitely proved beyond all peradventure that these fungous threads are the actual cause of the disease and that nothing else can cause this disease.

This fungous mycelium, coming in contact with a lettuce leaf, exudes a poison which kills the near-by cells of the lettuce plant. The cell walls are then dissolved and the mycelium makes its way through or between them. It dissolves also the contents of the cells and absorbs the resulting nutrient solutions to further its own development. The mycelium thus grows larger, kills more cells, consumes them and continues to advance rapidly through the affected leaf until the whole leaf is a soft, slimy, rotten mass. The invasion continues into the main stem, then upward to the central bud and "heart" of the head, out into other leaves, downward through the root until every portion of the host plant has been killed, and the nutritious parts consumed.

Environmental conditions may, to some extent, change the course of the disease. The fungus grows best in abundant moisture. Sometimes this leads to a more rapid decay of the inner protected dry portion of the head, and a plant which to casual inspection appears healthy may prove upon close examination to be, at heart, a slimy, rotten mass. Again through one-sided infection the decay may progress much more rapidly upon one side than upon another, resulting in complete death of one side before the other shows any symptom of disease.

As a rule the mycelium is not visible to the naked eye on leaves until the nutriment within the leaves has been nearly or quite, exhausted by the fungus. When this time is reached the mycelium begins its external appearance as the loose cottony growth referred to above. The most profuse development of this aerial mycelium occurs in the region of more humid atmosphere, such for example as on the under sides of leaves lying upon the ground, between leaves, or at the bases of leaves, in fact anywhere that the air is so situated as to cause it to remain especially humid.

Soon after the appearance of the aerial mycelium in profuse quantity, it may be noted that in each region where the mycelium is dense there appear one or more centers of aggregation, composed of very densely intertangled and interwoven mycelial threads. These denser masses enlarge, increase in density and finally become solid masses of tightly compacted mycelium. These bodies are called *sclerotia.* The sclerotia are at first colorless, then pale salmon color, and finally black on their exterior and flesh colored within. When first formed they are buried in and covered by mycelium, and are only to be seen by tearing this mycelial covering away. As time passes this mycelium is lost, the remains of the lettuce plant disappear, and the only visible

evidence of the plant or the fungus then is the sclerotia, many of which are produced in each sick plant.

The sclerotium germinates under suitable conditions, usually after a lapse of several months to nearly a year, under field conditions. This it does by sending forth several thread-like sprouts about one-thirty-second of an inch in thickness. These sprouts expand at the end developing a horn-like or cornucopia-like disk (Fig. 5) called the apothe-

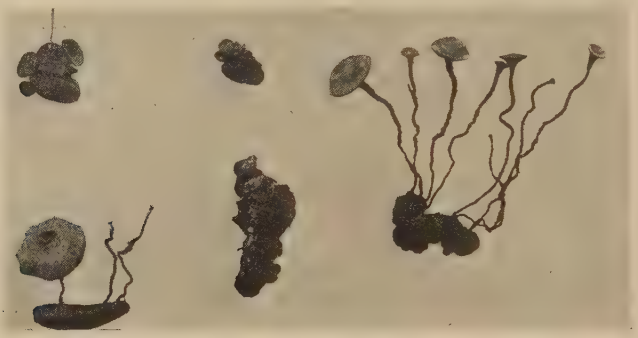


Fig. 5.—Sclerotia-bearing disks; natural size.

cium. This apothecium is, in the field, borne just at the surface of the ground with its face directed upward.

Microscopic examination of the apothecium shows its disks to consist of two parts: (1) lower basal part supporting (2) an upper layer which consists of very numerous small slender tubular bags, or sacs, the *asci.* (Fig. 6.) Each ascus when mature contains eight small

Fig. 6.—Asci and paraphyses in various stages of development.

oval bodies (Fig. 6) the *spores.* It is seen then that the apothecium is essentially an organ whose function is to produce myriads of spores.

When ripe these spores are ejected from the asci by pressure, being forced into the air often to a distance of several feet where, caught

by air currents, they may be carried quickly to great distances. The spores under suitable surroundings, germinate and send forth small thread-like sprouts (Fig. 7). These sprouts, with suitable nourishing material, grow rapidly into a vigorous mycelium, which is again ready to invade the living lettuce plant; to again cause the drop.

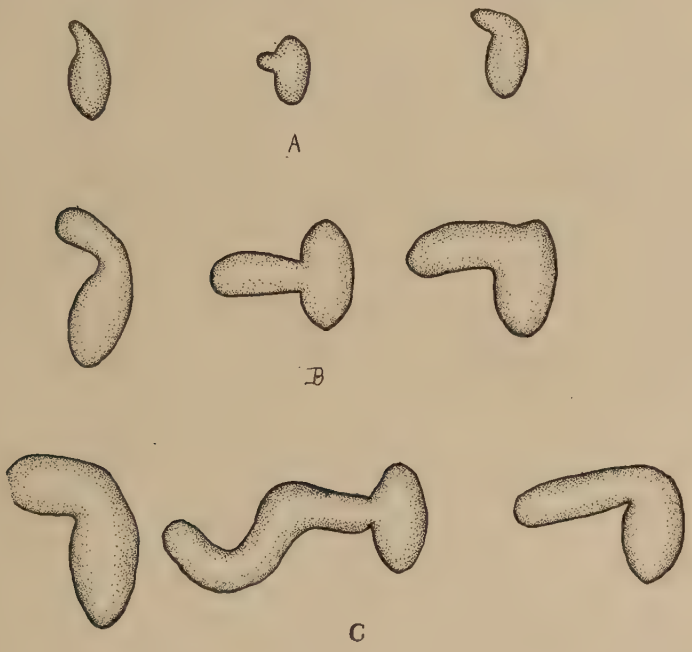


FIG. 7.—Ascospores germinating. *A*. At two hours; *B*, at five hours; *C*, at seven hours.

This history of the drop fungus may be epitomized by saying that the mycelium grows within the lettuce plant, causing the drop; produces sclerotia when nutriment is exhausted; rests in the sclerotial condition until opportune conditions prevail; the sclerotia then produce apothecia bearing asci in which are spores; these spores produce a new mycelium which again invades lettuce plants.

THE FUNGUS THAT CAUSES THE DISEASE.

MORPHOLOGY.

The *mycelium* in gross, as developed external to the leaf in humid air, appears as a coarse cottony mass most abundant around the stem of the plant among the leaf bases. It is also found to a great extent upon the lower diseased leaves that lie upon the surface of the ground. It is more plentiful on the under sides of these leaves or between leaves if two or more are lying upon each other on the ground, but very seldom makes any extensive showing upon the ground itself unless under artificial conditions of excessive humidity.

The mycelium is comparatively large and coarse, varying in diameter from 5.8 μ to 14.5 μ with many septa (Fig. 8) which divide each mycelial thread into many cells, whose lengths vary with their position

FIG. 8.—Mycelium showing septation and branching.

FIG. 9.—Mycelial thread, showing mode of apical branching.

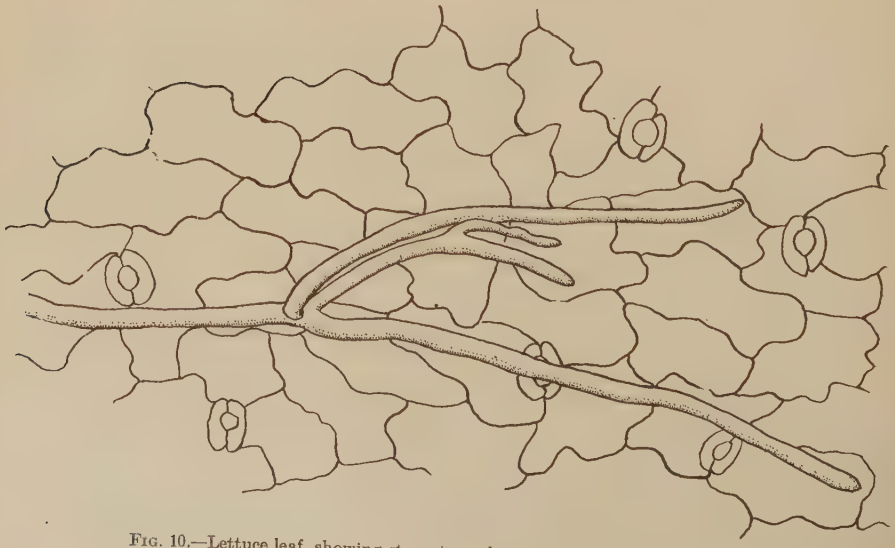


FIG. 10.—Lettuce leaf, showing stomata and superficial sclerotinia mycelium.

in the hypha. In the younger growing tips the septa are very far apart, and become closer in the older parts. There is no completely regular method of hyphal branching, although the greater number of

FIG. 11.—Mycelial thread showing three stages in its development; *a*, young; *b*, medium; *c*, old.

branches seems to arise as perpendicular outgrowths near the end or middle of a cell, Fig. 8. There are two other methods of branching, one of which will be discussed under "attachments." The other is

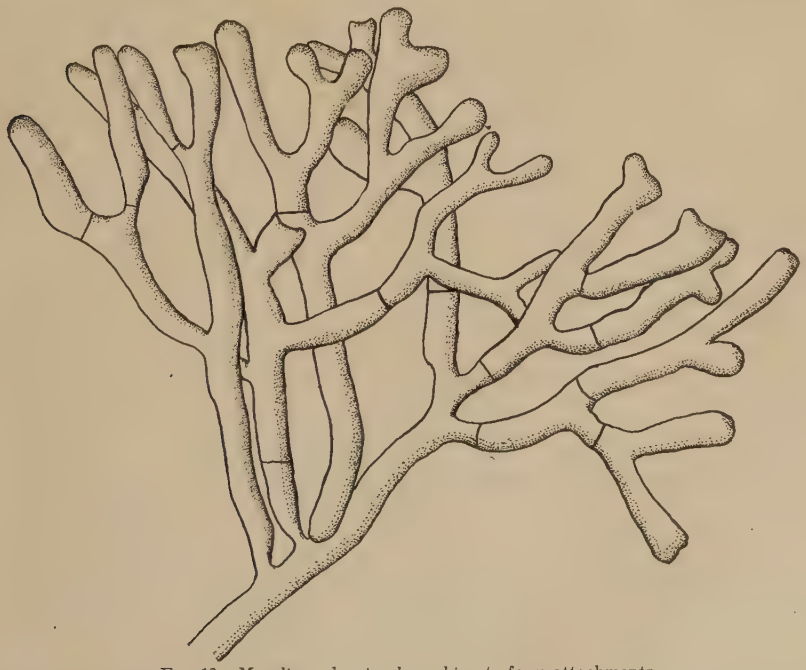


FIG. 12.—Mycelium showing branching to form attachments.

that in the case of a young hypha, which is growing rapidly and has an abundance of nourishment, in which case three or more branches arise, frequently simultaneously, and the main hypha loses its predominance; the three branches being equally vigorous. At first,

as in the youngest mycelium, the contents of each thread is a more or less homogenous mass, Fig. 11a, of protoplasm, but gradually as the hypha grows, there appears first a single row of vacuoles,

Fig. 13.—Portion of attachment organ showing flattening of the tips.

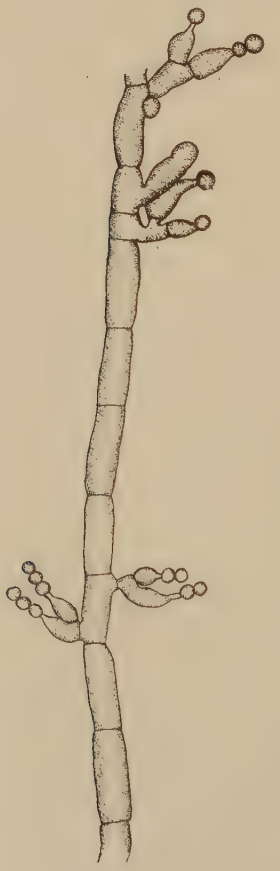


Fig. 14.—Mycelium showing method of production of gonidia.

Fig. 11b. These increase until the whole filament becomes very vacuolate, and finally all apparently coalesce to one large vacuole which nearly fills the whole of the cell, Fig. 11c.

The *attachments* first appear as numerous short branches from near the tip of an ordinary hypha. Each one of these primary off-shoots produces branches that again branch forming a rather compact mass, Fig. 12. They are at first continuous but very soon become septate.

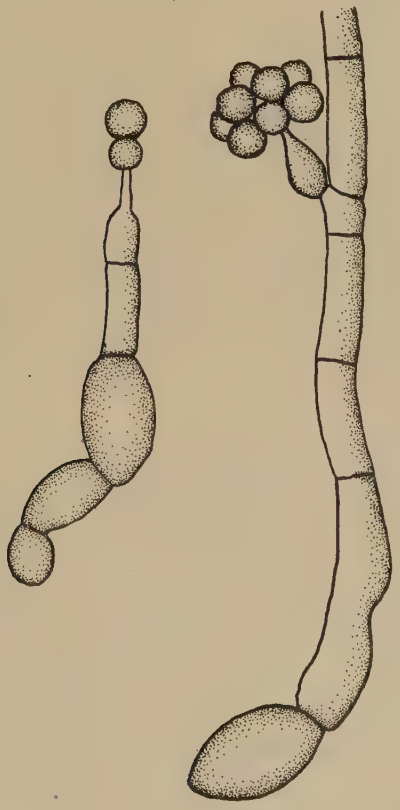


Fig. 15.—Germ tubes from ascospores producing gonidia.

With the appearance of the septa the tips of the branches enlarge. When they come into contact with some solid as a culture dish or a lettuce leaf, they become somewhat flattened, Fig. 13, on the end and apply themselves to and cohert to the substance.

Gonidia.—Frequently both in drop and in plate cultures there are found small spherical bodies, *gonidia,* that appear very much like spores, Fig. 14, although they have never been seen to germinate. They are highly refractive bodies, measuring about 2 to 3 mu in diameter, and are produced in large numbers in acropetal succession upon flask-shaped stalks. These stalks are produced either as lateral branches of an ordinary vegetative hypha or as the termination of

such hypha. They are at times also produced in abundance upon the germ tubes formed by the ascospore, Fig. 15. In some cases several are formed close together, so that many gonidia are produced together, forming a large mass and completely hiding the stalks that bear them.

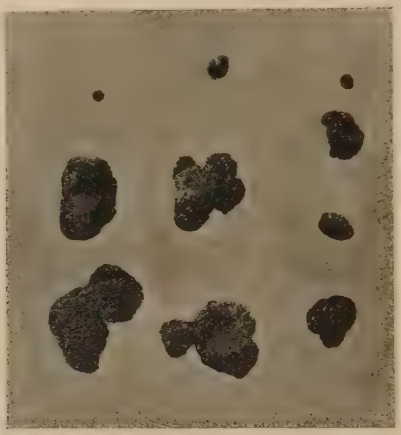


FIG. 16.—Sclerotia taken from one infected plant, showing variation in size.

Sclerotia.—Upon suitable media in the laboratory and upon plants in the field the purplish black resting bodies, the sclerotia, are formed. These vary much in shape, some of the smaller ones being almost

FIG. 17.—Section of a sclerotium showing parenchyma-like rind and modified mycelium of the interior.

spherical and some of the larger ones long, irregularly cylindrical or flat. In size there is also much variation, the smallest ones being no larger than a pin head, while the larger ones are larger than a grain

of corn. The large and the small are however in nature and in normal cultures, produced indiscriminately together, and in no case was there a production of either large or small ones exclusively.

When first gathered from the field their average weight was determined as .06 grams each. After dying in the laboratory for a year they averaged .02 grams as determined upon a weighed hundred. They are somewhat rough on the surface and have a rather tough exterior protecting layer composed of two or three layers of parenchyma-like cells with thick, hard, black walls, Fig. 17. Internally the sclerotium consists of a compacted mass of mycelial threads, which do not vary perceptibly from the ordinary mycelium, except that they have much thicker walls and narrower lumen, Fig. 17, and their diameter is less than that of the vegetative hyphæ.

In cultures in the laboratory it was possible to follow the development of the sclerotia accurately. At first there occurs a massing of the mycelium, causing a white, somewhat raised bunch of mycelial threads, floccose in appearance. These masses as they grow, exude numerous small drops of a colorless watery liquid, after which they change to cream color, which gradually passes to a dirty yellow, the developing sclerotial membrane losing their floccose character. Their color changes to a greenish black, afterward to almost pure black as they mature.

FIG. 18.—Sclerotium with disks. *a*, Photographed January 10; *b*, January 12; *c*, January 14. Note the recurved edges of the largest disk.

To secure *germination of the sclerotia,* they were placed in moist sand in flower pots which were kept standing in a pan of water thus keeping the sand wet by capillarity. After about two months the sclerotia began to send out minute yellowish-brown bud-like protuberances. These rapidly elongated to form a filament which as soon as it reached above the soil and into the light, began to expand, at first to an urn-shape and then gradually flattening into a broad, flat disk, Fig. 18. In some cases the disk became recurved with age, Fig. 17c, having the appearance of a very minute yellowish brown umbrella,

the ascophore. From 3 to 35 ascophores were produced from each sclerotium.

The Ascophore.—The *Ascophore* may be regarded as composed of two portions, the stalk or stipe, and the disk, though no sharp line of demarkation can be drawn between these parts. As many as thirty-five stipes have been noted from a single sclerotium, but usually there are not more than eight or ten. There are no particular points upon the sclerotium from which they arise, but in case the sclerotium lies upon the surface of the ground, they usually spring from the sides of it nearest to the soil. Their length depends upon the depth of the sclerotium in the soil. If the sclerotium is upon the soil surface, the stipe is just long enough to allow the disk to expand. When the sclerotium is buried the stipe becomes a sufficient length to reach the surface, sometimes attaining a length of 3 to 5 cm. In thickness the stipes vary from about 0.5 to 1.2 mm. The color of the shorter stipes is almost universally brownish yellow, but the longer ones while brownish yellow just below the disk are dark brown to almost black at the sclerotial end.

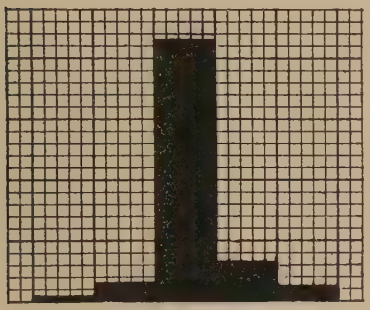
In cross section the stipe is seen to be hollow and to possess an outer cortical region composed of two or three irregular layers of black thick-walled cells, encasing a tissue of parenchymatous appearance. In longitudinal section, cells of the outer cortical layers appear about three times as long as wide, while the rest of the tissue appears as a mass of ordinary mycelial threads with rather more than the usual number of septa.

The disk is borne upon the end of the stipe. It is at first very small and deeply cup shaped. This cup gradually expands at the rate of one-half to 1 mm. per day and forms the flat or concave circular disk, which sometimes attains a diameter of 16 mm. It is composed of two distinct parts, the *stroma* and the *hymenium*. The stroma or basal part consists of a mass of closely interwoven hyphæ with an outer cortical parenchymatous layer two or three cells thick which forms the under surface of the disk. The stroma supports the hymenium composed of *asci* (Fig. 6) and *sterile* hyphæ lying between them, the paraphyses (Fig. 6). The asci are cylindrical with a gradually narrowing base. They measure about 82 x 2 mu and bear 8 spores in their distal half. The asci are very numerous, there being about 30,000 in a single disk of usual size. Between the asci and more numerous than the asci are the very fine thread-like paraphyses which are slightly longer than the asci, and not more than one-third as thick. The paraphyses do not differ materially from the ordinary hyphæ of the mycellium.

The spores found in the asci are hyaline, oblong, elliptical with somewhat pointed ends, and when mature bear two vacuoles. They measure 5.8 x 8.7-11.6 mu.

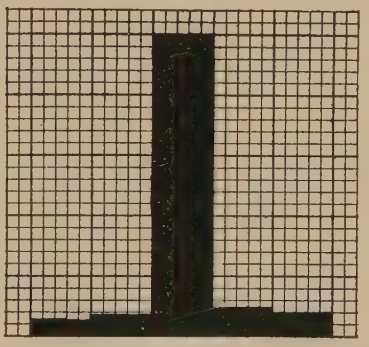
In a single disk there are about thirty-one million spores; in a single sclerotium of average size about three hundred and ten million spores. Allowing a fair number of sclerotia to each diseased lettuce plant it is seen that it can produce as many as five billion spores.

When mature the spores are ejected forcibly from the asci. Great numbers of them ripen at the same time and the slightest change in environmental conditions, as a slight draught produced by the breath causes myriads of them to be thrown into the air often to a distance of 0.5 to 1.0 meter, their great number rendering them visible as a steam-like cloud which, caught by air currents, can often be followed by the eye to a distance of several meters.



$M = 4.0880 \pm 0.0166$
$\sigma = 0.2930 \pm 0.0117$
C. V. $= 7.168 \pm 0.290$
$n = 142$

FIG. 19.—*Sclerotinia libertiana* Fckl. Polygon of ascospores from middle-aged disk.



$M = 4.0393 \pm 0.0214$
$\sigma = 0.3743 \pm 0.0151$
C. V. $= 9.267 \pm 0.380$
$n = 165$

FIG. 20.—*Sclerotinia libertiana* Fckl. Polygon of spores from young disk.

Several hundred measurements were made from disks of various ages to determine the normal variation in sizes of ascospores with the results shown in Figs. 19 and 20.

It is seen that the spores are remarkably constant in size and that the age of the disk (Figs. 19 and 20) from which they are taken has no material effect upon their measurements.

The spores as discharged from the asci are ready, if they fall in favorable places, to germinate at once. In germination they first en-

large somewhat and soon a protuberance can be seen at some spot on the surface. This protuberance increases in length, becomes septate and constitutes the germ tube, the beginning of the mycelium.

The Species of the Fungus.

The species under consideration has by recent writers been regarded as *Sclerotinia libertiana* Fuckel which is thus described[19]:

"Sparsa, stipitata, nuda, pallida, cyathoidea, 4-8 m.m. lat.; stipite tenui subflexuoso, *Sclerotio* tuberiformi, nigro innato, plus minus elongato, sæpissime 3 cm. long; ascis cylindraceis, 130-135 x 8-10, apice jodo dilute coerulescentibus; sporidiis monostichis, ellipsoideis, vulgo minute güttulatis, 9-13 x 4-6.5; paraphysibus parcis, clavulatis."

Comparison of the fungus under discussion with the description of eleven species which most nearly agreed with it showed that all failed to agree with it in some important point except *Sclerotinia libertiana* and *S. kauffmanniana;* the latter considered a synonym of *S. libertiana* by some authors.

Physiology.

The Mycelium.

Media.—This fungus lends itself readily to culture upon either artificial or live media. Its growth upon various media was first studied to ascertain the best medium to employ in experimental work and the best medium for stock cultures.

Upon lettuce leaves in culture dishes a great mass of mycelium of loose cottony consistence, which very quickly collapsed on exposure to the air, was produced and very few sclerotia were formed.

In lettuce broth a vigorous mycelium developed.

Lettuce agar gave only a very thin surface layer of mycelium, and very few sclerotia.

Four-per-cent. pea-agar gave about the same growth as did lettuce agar.

Sterilized corn meal, wet with apple juice, gave excellent growth and produced by far the greatest number of sclerotia. The mycelium while mainly coating the surface of this medium was very compact and dense. Corn meal wet with lettuce broth was equally satisfactory. In all of the foregoing media a much more abundant mycelium was formed if the medium was slightly acid.

The corn-meal-apple-juice mixture was adopted for the study of the development of sclerotia and as a medium on which to maintain stock cultures. Lettuce broth was used to grow mycelium for tests regarding the effects of soluble chemicals, since the mycelium developed well, and uniformly, in this medium and was readily wetted by the solutions employed in the experiments which was not the case when media leading to the development of a profuse aereal mycelium were employed.

Experiment 9, Inoculated 11-30-1906. To determine the *temperature relations* of the fungus.

Sclerotinia libertiana was grown upon lettuce agar, acidity +.55 Fuller's scale, and upon lettuce leaves at cool, room temperature, incubator temperature and in an open shaded window, all in ordinary diffused light except the culture in the incubator room which were in the dark. The results are shown in the following table.

TABLE I.—SHOWING RELATION OF TEMPERATURE TO GROWTH.

FIGURES INDICATE TOTAL GROWTH RECORDED AS MILLIMETERS.

Medium	Condition	Growth										
		December										
		1st	3d	4th	5th	6th	7th	8th	10th	11th	12th	13th
Agar	Window shaded	1	5	5	5	10	15	17	23	30	35	37
Lettuce leaf	Window shaded	0	0	5	5	25	30	40	60	60	70	70
Agar	Cool room	4	13	20	40	40	47	53	70	70	78	------
Lettuce leaf	Cool room	20	20	20	35	50	------	------	------	------	------	------
Agar	Incubator room	4	6	6	6	6	6	6	6	6	6	------
Lettuce leaf	Incubator room	4	4	4	------	------	------	------	------	------	------	------

It is clearly evident that the incubator temperature (approximately 37 1-2 degrees) was unfavorable to the growth of the fungus. Growth stopped entirely after the first few millimeters and was not resumed, nor were any sclerotia formed.

The room temperature, which was considerably warmer than the window temperature of December, was more favorable to growth, resulting in complete occupancy of the plate in 9 to 11 days. While growth was slower at the colder outdoor temperature it still continued vigorously and normally, leading eventually to complete occupancy of the plate. Sclerotia were formed upon the lettuce leaf at both room and outdoor temperatures.

Experiment 48, March 12, 1908. To determine more accurately the optimum growth temperature in agar, cultures were made in this medium and placed in an optimum temperature apparatus.*

The work was done in quadruplicate and care was taken to have all cultures as nearly as possible parallel as to moisture, quantity and quality of inoculum and all factors except temperature. The results are expressed in table II.

It is seen that growth was markedly less in the two warmer compartments (29 and 36-39 degrees). No growth at all occurred in the warmest compartment and no sclerotia formed in either of these compartments. In the compartment of lowest temperature (13-18 degrees) growth was also much retarded but it still continued, leading to complete occupancy of the plate in twelve days and to sclerotial formation in thirteen days.

*An incubator with six compartments, giving a considerable range of temperatures.

TABLE II—SHOWING RELATION OF TEMPERATURE AND GROWTH.

	Temperature—Compartment Number								‡ Growth—Compartment Number					
	1		2	3	4	5	6		1	2	3	4	5	6
Date	Max.	Min.					Max.	Min.						
3-14-08	13	16	21	24	26	29	36	39	0	4	5	3	4	0
3-16-08	13	21	23	25	27	29	36	39	10–15	16–21	20	17	16	0
3-17-08	13	23	21	24	26	29	36	39	0–10	13–15	13	10–15	7	0
3-18-08	13	17	21	24	26	29	36	39	5–10	13–16	14–17	0– 5	8	0
3-19-08	14	17	21	24	26	29	36	39	7–27	13–21	5– 9	2	5	0
3-20-08	13	16	21	24	26	29	36	39	8–25	F*	F	8	5	0
3-21-08	14	17	21	24	26	29	36	39	5–10	F	F	10	5	0
3-23-08	13	18	21	24	26	29	36	39	10	F	F	10	10	0
3-24-08	13	16	21	24	26	29	36	39	10–25	F	F	F	0	0
3-25-08	12	18	21	24	26	29	37	39	F	S†	S	S	0	0
3-26-08	13	18	21	24	26	29	36	39	S	S	S	S	0	0
Average number of sclerotia per culture									1	2	2	1.8	0	0

* F indicates date upon which mycelium filled plate.

† S indicates that sclerotia were forming.

‡ Growth for each day is recorded in millimeters. In compartments 1 and 6 maximum and minimum thermometers were used.

Of the four compartments of intermediate temperature the two cooler (21 and 24 degrees) were more favorable than the two warmer (26 and 29 degrees). In the two former complete occupancy of the plate by the mycelium occurred in six days while the others required four days more.

From these tests we may conclude that this fungus can not continue to grow long at a temperature of 29 degrees or higher; that at a temperature as low as 13-18 degrees or as high as 26 degrees growth proceeds normally but not so rapidly as at the optimum temperature; which lies between 21 and 24 degrees.

Experiment 13. To determine *longevity of mycelium* in agar in petri dishes. A ten-per-cent. lettuce agar was employed, acidity +.55 Fuller's scale. Tests were made in the laboratory, the results of which appear in table III.

From this table it is seen that the mycelium remained alive as long as forty-eight days, but in no case did it live beyond the fifty-fifth day. The length of life after the agar was dry in the petri dish varied from five to ten days.

Experiment 12. To determine the *effects of various nutrients* upon growth. Sclerotinia was grown in petri dishes upon plain agar with various nutrients added. 10 c.c. of medium was used in each dish. The petri dishes were inoculated 12-11-1906.

TABLE III.—SHOWING LONGEVITY OF MYCELIUM IN AGAR IN PETRI DISHES.

Plate No.	Date of Inoc.	Date of Filling Plate	Date of Drying	tested	alive	tested	alive	tested	alive	tested	alive	tested	alive	tested	alive
1	5–15–09	5–21	6–20	6–1	Yes	6–14	Yes	6–23	Yes	6–30	Yes	7– 7	No	------	------
2	5–15–09	5–21	6–20	6–1	"	6– 8	"	6–14	"	6–23	"	6–30	Yes	7– 7	No
3	5–15–09	5–23	6–22	6–1	"	6– 8	"	6–14	"	6–23	"	6–30	"	7– 7	No
1	6–14–09	6–18	7–18	6–18	"	7– 7	"	7–14	"	7–23	"	8– 3	No	------	------
2	6–14–09	6–18	7–18	6–18	"	7– 7	"	7–14	"	7–23	"	8– 3	"	------	------
3	6–14–09	6–18	7–18	6–18	"	7– 7	"	7–14	"	7–23	"	8– 3	"	------	------
4	6–14–09	6–18	7–18	6–18	"	7– 7	"	7–14	"	7–23	"	8– 3	"	------	------
1	9–23–09	9–27	10–28	9–30	"	10–14	"	10–28	"	11– 7	"	11–10	"	------	------
2	9–23–09	9–27	10–28	9–30	"	10–14	"	10–28	"	11– 3	"	11–10	"	------	------
3	9–23–09	9–27	10–28	9–30	"	10–14	"	10–28	"	11– 3	"	11–10	"	------	------

As might have been expected growth was very slow in plain agar, while 5 per cent. glucose, and 1 per cent. starch proved most favorable. Lettuce agar was much superior to plain agar but far less nutritious than starch or glucose agars.

The deficiency of the peptone and lactose agars is striking, seeming to emphasize the lack of need of rich nitrogenous foods for this fungus and to show the superior value of glucose over lactose as a source of carbon. There was no reddening of the litmus lactose agar by the mycelium.

Sclerotia were formed on all of these media except the peptone and the litmus lactose agar.

TABLE IV.—SHOWING EFFECTS OF VARIOUS NUTRIENTS UPON GROWTH.

GROWTH IS RECORDED IN MILLIMETERS.

	Medium	Days									
		1	2	3	4	5	6	7	8	9	10
1	Plain agar	5	9	14	20	23	25	30	30	39	---
2	Plain agar	2	2	12	16	25	32	35	36	43	---
3	Plain agar+1 per cent glucose	3	11	25	28	34	38	42	----	----	---
4	Plain agar+1 per cent glucose	1	4	10	10	15	24	26	----	----	---
5	Plain agar+5 per cent glucose	2	8	40	65	----	----	----	----	----	---
6	Plain agar+5 per cent glucose	3	25	75	Fills Plate	----	----	----	----	----	---
7	Plain agar +1 per cent peptone	2	5	10	10	12	12	15	----	----	---
9	Plain agar+Litmus lactose	2	10	20	24	26	28	----	----	----	---
10	1 per cent starch agar	3	13	30	40	45	50	60	----	----	---
11	1 per cent starch agar	2	8	25	42	----	50	58	----	----	---
12	Lettuce agar	4	13	20	40	47	53	70	----	----	---
13	Lettuce agar	4	8	14	22	32	42	48	----	----	---

Experiment 8. To determine the *influence of acidity or alkalinity* of the medium. On December 8, 1906, the fungus was inoculated and grown in lettuce agar of various degrees of acidity and alkalinity as is shown in the table following.

TABLE V—SHOWING RELATION OF ACIDITY TO GROWTH.

Total Growth Is Recorded in Millimeters.

Drops of Normal HcL or NaOH	Fuller's Scale	Day									
		1st	2nd	3rd	4th	5th	6th	7th	8th	9th	10th
+20											
+15											
+10	+40.55	0	0	0	0	0	0	0	0	0	0
+5	+20.55	0	2	3	5	7	12	16	16	18	18
0	+ .55	6	14	28	36	41	67	75	83	83	83
—5	—19.45	0	10	0	0	0	0	0	0	0	0
—10	—39.45	0	0	0	0	0	0	0	0	0	0
—15	—59.45	0	0	0	0	0	0	0	0	0	0
—20	—79.45	0	0	0	0	0	0	0	0	0	0
Date	December	11	12	13	14	15	17	18	19	20	21

It is seen that growth was inhibited by + 40.55 and — 19.45 of Fuller's scale. At + 20.55 growth was slow. At + 0.55 occurred the maximum growth.

Experiment 15. To determine the *toxicity of various fungicides* acting directly upon the mycelium.

Sclerotinia was grown in glass capsules in lettuce broth until a vigorous mycelium was obtained. The broth was then poured off and the mycelium was washed free of any remaining broth by allowing about one litre of sterile water to pass slowly through it by means of a small siphon. A small portion of the mycelium, the quantity as nearly equal as possible in all cases, was then transferred to the poison, the effect of which was to be tested. After ten minutes, one-third of this mycelium was removed, rinsed in several changes of sterile, distilled water and placed in tubes of sterile lettuce agar to test its viability. Other portions of the mycelium were similarly removed at the end of an hour and of twenty-four hours. The temperature of the poisons when used was approximately 21 degrees. The results are shown in table VI.

This experiment shows the inefficiency of a ten minute's application of such fungicides as ammoniacal-copper-carbonate, strong and weak Bordeaux, formalin 1 ounce to 1 gallon, and 1 ounce to 2 gallons, saturated lime water, and potassium permanganate 1 ounce to 10 gallons, though each of the above is fatal if applied for one hour.

TABLE VI.—SHOWING TOXICITY OF VARIOUS FUNGICIDES.

+ = viable; — = not viable.

Fungicide	Strength	10 Min.	1 Hr.	24 Hrs.
Ammoniacal Copper Carbonate	Copper Carbonate, 6 ozs.; Ammonia, 3 pts.; H_2O 50 gals.	+	—	—
Bordeaux Mixture	5 lbs. $CuSO_4$, 5 lbs. Lime, 50 gals. H_2O	+	—	—
Bordeaux Mixture	2 lbs. $CuSO_4$, 2 lbs. Lime, 50 gals. H_2O	+	—	—
Potassium Sulphide	1 oz. Potassium Sulphide, 1 gal. H_2O	+	+	+
Potassium Sulphide	1 oz. Potassium Sulphide, 3 gals. H_2O	+	+	+
Lead Acetate	1 oz. Lead Acetate, 1 gal. H_2O	+	+	+
Lead Acetate	4 ozs. Lead Acetate, 1 gal. H_2O	+	+	+
Potassium Permanganate	1 oz. Potassium Permanganate, 1 gals. H_2O	—	—	—
Potassium Permanganate	1 oz. Potassium Permanganate, 3 gals. H_2O	—	—	—
Potassium Permanganate	1 oz. Potassium Permanganate, 5 gals. H_2O	—	—	—
Potassium Permanganate	1 oz. Potassium Permanganate, 7 gals. H_2O	—	—	—
Potassium Permanganate	1 oz. Potassium Permanganate, 10 gals. H_2O	+	—	—
Formalin	1 oz. Formalin (40 %), 1 gal. H_2O	+	—	—
Formalin	1 oz. Formalin (40 %), 2 gals. H_2O	+	—	—
Formalin	1 oz. Formalin (40 %), 3 gals. H_2O	+	+	+
Sodium Benzoate	1 oz. Sodium Benzoate, 1 gal. H_2O	+	+	+
Sodium Benzoate	1 oz. Sodium Benzotte, 2 gals. H_2O	+	+	+
Sodium Benzoate	1 oz. Sodium Benzoate, 5 gals. H_2O	+	+	+
Sodium Benzoate-Bordeaux	½ lb. Sodium Benzoate, 1 lb. $CuSO_4$, 1 lb. Lime, 50 gals. H_2O	—	—	—
Iron Sulphate	15 %	+	+	+
Lime Water	Saturated	+	—	—
Lime Paste		—	—	—

Potassium sulphide, 1 ounce to 3 gallons and 1 ounce to 1 gallon, lead acetate 1 ounce to 1 gallon and 4 ounces to 1 gallon, formalin 1 ounce to 3 gallons, sodium benzoate 1 ounce to 1 gallon, 1 ounce to 2 gallons, 1 ounce to 5 gallons, and iron sulphate were not fatal even with a twenty-four-hour application.

Potassium permanganate 1 ounce to 1 gallon, 1 ounce to 3 gallons, 1 ounce to 5 gallons, and 1 ounce to 7 gallons, lime paste and sodium-benzoate-Bordeaux (one-half pound sodium benzoate, 1 pound copper sulphate, 1 pound lime to 50 gallons) were fatal even with the shortest period tested.

The inefficiency in these comparatively strong solutions of potassium sulphide and of formalin indicates considerable vigor and resistance on the part of the fungus.

Experiment 19. Copper sulphate was tested following the method of the last experiment, using strengths varying from N/100 to N/6400 and varying the time from 5 to 105 minutes. With the three weaker strengths, tests were made continuing to twenty-four hours. The tests were made with the solutions at a temperature of 17 degrees. The results are shown in the following table:

TABLE VII.—SHOWING TOXICITY OF COPPER SULPHATE.

o = Not viable; + = Viable.

Strength of Solution.	Time in Hours.																								
	1/12	1/6	1/4	1/3	5/12	1/2	7/12	2/3	3/4	5/6	11/12	1	1 1/12	1 1/6	1 1/4	1 1/3	1 5/12	1 1/2	1 7/12	1 2/3	1 3/4	4	8	24
n/100	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	o	o	o	o	o	o	o	o
n/200	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	o	o	o	o	o	o	o	o
n/400	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	o	o	o	o
n/800	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	o	o	o	o
n/1600	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
n/3200	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
n/6400	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+

In all cases here copper sulphate failed to cause death of the mycelium, until the mycelium had been subjected to the solution for 80 minutes. It then caused death with the two stronger solutions, N/100 and N/200. N/400 and N/800 did not cause death till the expiration of 105 minutes, and even 24 hours soaking in the three weaker solutions, N/1600, N/3200 and N/6400, failed to kill the fungus.

In conditions unfavorable to mycelial growth, for example upon plain agar, or agar otherwise deficient in nutrients the gonidia were produced in abundance. Their production also occurred in the midst of dense masses of mycelium in late periods, apparently due to the unfavorable effects of lack of nutrients or to the development of inhibiting products. They were also formed abundantly from the germ tubes of ascospores in drop cultures in sterile water within a few days after the cultures were made. Here no nutriment was accessible except that present in the spore itself.

Careful count was made of the number of sclerotia upon various parts of affected lettuce plants. The results are given in Table IX.

These plants were naturally infected in the lettuce beds. It is seen that the greater number of sclerotia was formed in the axils of the leaves around the stem. Those formed in the ground on the root were produced only after the root had begun to decay. No plants that were examined, in which the roots had not begun to decay, showed signs of sclerotia below ground and in very few instances was there any external sign of the fungus in the parts below ground.

If half of a bed of the usual size, 9 x 100 feet, bearing 2,000 plants, be diseased (this is not an uncommon percentage), it is seen that there may be formed as many as 17,100 sclerotia per bed. It is quite

certain that this estimate does not fully represent the actual number formed under conditions of badly infected beds.

TABLE VIII.—SHOWING DISTRIBUTION OF SCLEROTIA IN THE PLANT.

Plant No.	On Ground	In Axils of Leaves	Among Leaves	On Ground in Root	Total
1		9	2	1	12
2		9		2	11
3		9			9
4	1	14	8	10	33
5		9	1	4	14
6		13	2	4	19
7		8	14	4	26
8		23	1		24
9		10		4	14
10		9			9
Total	1	113	28	29	171
Average	1	11.3	2.8	2.9	17.1

Sclerotia can be grown upon a great variety of media but upon some media they do not develop in any large quantity. They were formed upon the surface of the lettuce broth cultures after the mycelium had grown to a considerable extent, thus forming a support for them; but sclerotia have never been seen to form within the liquid medium itself and plants brought into the laboratory and kept in culture dishes where they became very wet did not form as many sclerotia as those placed upon soil in the greenhouse where the plants were less wet.

In a test of plain agar, plain agar plus 1 per cent. glucose, plain agar plus 5 per cent. glucose, plain agar plus 1 per cent. peptone, plain agar plus litmus-lactose, plain agar plus 1 per cent. starch and lettuce agar, it was seen that sclerotia formed in all the media except those which contained peptone or litmus-lactose. In a test of temperature relations no sclerotia developed in the higher temperatures, above twenty-six degrees C.; nor at the lower temperatures, below eight degrees C. The formation of sclerotia is inhibited by these extremes before the growth of the mycelium is stopped.

Two cultures of Sclerotinia upon the corn meal saturated with apple juice, in 20 cm. culture dishes, were inoculated January 17, 1907, and the first mature sclerotia were picked from these cultures on January 28th, 11 days from inoculation.

From these cultures full size, black sclerotia were taken every alternate day until February 8th, then every third day until February 17th; a period in all of one month from the date of inoculation. During this time one of the cultures produced 523 sclerotia and the other 397 or a total of 920 for the two cultures. The following table shows the number of sclerotia taken on each date.

TABLE IX.—SHOWING SCLEROTIA PRODUCTION IN CULTURES.

Date	Jan. 28, '07	Jan. 30, '07	Feb. 2, '07	Feb. 4, '07	Feb. 6, '07	Feb. 8, '07	Feb. 11, '07	Feb. 14, '07	Feb. 17, '07	Total
Sclerotia taken from Culture, A	52	39	105	10	39	75	67	65	71	523
Sclerotia taken from Culture, B	52	89	39	75	10	41	29	26	36	397
Total	104	128	144	85	49	116	96	91	107	920

The formation of sclerotia seems dependent upon the attainment of a certain mycelial density. Thus upon a rich medium, as corn meal, continued branching accompanied by radial enlargement of the colony

FIG. 21.—Sclerotia produced in culture upon corn meal wetted with apple broth, showing concentric rings of sclerotia around the central point of inoculation.

soon leads to mycelial crowding. A quite regular ring or zone of sclerotia results, Fig. 21. Again radial expansion begins and after a certain interval another ring of sclerotia is produced. This alternation is repeated indefinitely.

When several colonies are so located that they meet mycelial crowding must occur at the points of contact. Here, too, sclerotia develop in

abundance, Fig. 22. Attention has been called elsewhere,[18] to the importance of mycelial crowding as a stimulus.

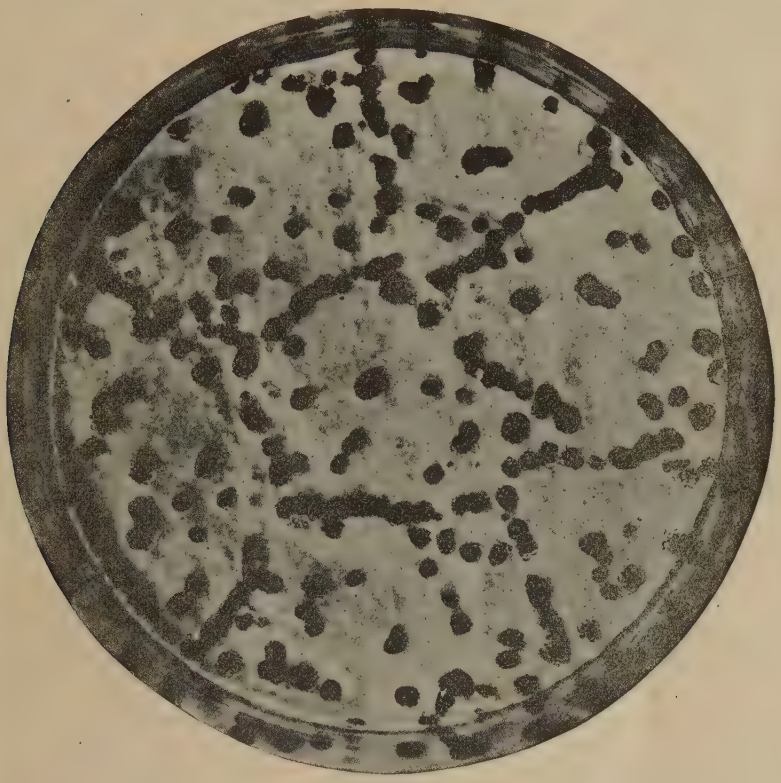


FIG. 22.—Sclerotia produced in culture upon corn meal wetted with apple broth, showing rows of sclerotia where colonies met.

To determine the length of time necessary for sclerotia to mature, for germination, a large number of sclerotia was taken from corn meal cultures and air dried for a few days. They were then planted in pots of sterilized soil at periods of a week apart. At each of these plantings old sclerotia, that were known to be mature, were also planted as controls. All of these grew, even those which were picked and merely air dried for a day or so showing as vigorous germination as did the old sclerotia. It thus appears that sclerotia are mature as soon as formed and are ready to germinate at once if favorable conditions obtain.

Attempts were made to determine the length of life of sclerotia under laboratory conditions and lettuce bed conditions. In the laboratory sclerotia were stored in ordinary room temperature, one lot in the dark all the time, another lot alternately in the light and dark of day and night, a third lot was kept wet all the time. Three similar sets were kept in the incubator room (temperature 37 degrees). At the end of 11

months those kept wet were found to have all decayed. Of those kept dry a little more than one-third (24 out of 60) produced ascophores normally. In an out-door test a large quantity of sclerotia was collected, placed in the lettuce beds at one-half inch depth in March. These were examined twice during the early part of the summer and were all sound. The third examination was made August 11, 1909 (five months and eleven days after placing them in the soil) and it was found that they had all decayed and the only thing left was the hard, black, parenchymatous, shell-like outside coverings and a very few of the thread-like stumps of stipes, abortive attempts at germination. While the longevity of the sclerotia under conditions most favorable to them has not yet been determined, it is evident that *under usual conditions their number must be largely reduced by decay.*

Germination of Sclerotia.

To determine the conditions most favorable to their germination sclerotia were placed:

(1) In ordinary three-inch flower pots filled with soil and these placed in a shallow dish of water. The pots were covered with strips of bibulous paper with the ends bent down into the water. The sclerotia were pressed into the soil until they were just level with the surface.

(2) Similar pots were prepared in the same way and placed in the incubator room and covered with bell jars instead of paper.

(3) Pots were placed outdoors on the north side of the building in a shady and cool location.

(4) Pots prepared as in the first case but not covered with paper were kept in the laboratory taking precaution only to keep water in the dish in which they were placed.

Ascophores were produced in all cases except under the incubator room conditions in which case no ascophores were formed until the pots were removed to the laboratory. The fourth method was found to be the most satisfactory and convenient for laboratory use.

Apothecia produced under unequal illumination are strongly positively phototropic turning their disk faces toward the source of light.

An experiment was made to determine the maximum depth in the soil at which sclerotia would germinate and produce apothecia. Three six-inch flower pots were filled with soil and each pot was divided into two compartments by glass partitions, sclerotia were then planted at depths of 1-2, 1, 1 1-2, 2, 2 1-2, and 3 inches. No apothecia were formed from sclerotia placed at a greater depth than 1 1-2 inches and by far the largest number of apothecia was formed from sclerotia at the minimum of the above depths.

To study the effect of stirring the soil over the sclerotia when in conditions favorable to their germination sclerotia were planted in soil in a two-foot flat in the laboratory and kept properly watered. Half of the flat was left intact and undisturbed. In the other half the soil surface was stirred or hoed every two weeks. In the undis-

turbed half apothecia were formed normally. More than 10 were counted at 25 days after planting. In the half of the flat in (Fig. 23) which the soil was stirred, no apothecia at all were formed even after 60 days' test.

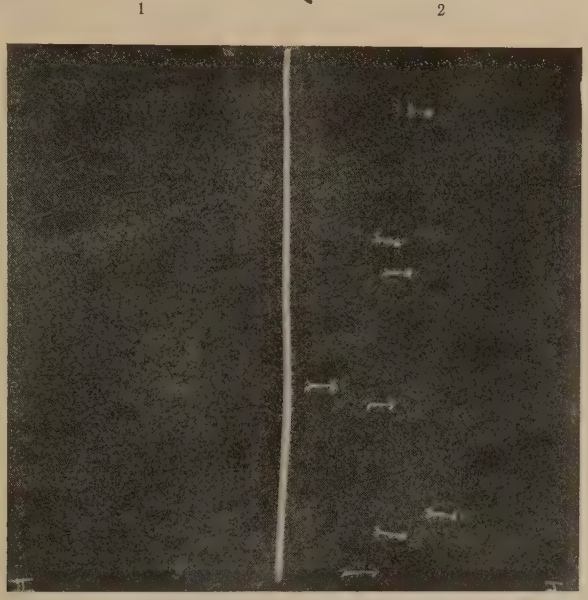


FIG. 23.—Flat showing the effect of stirring the soil upon the formation of apothecia. 1, Not stirred. 2, stirred every two weeks.

THE ASCOPHORE.

To determine the effect of mutilation of the ascophore, sclerotia were germinated and the stipes, as soon as they began to expand, were cut off at various distances from the apex. It was found that these stipes ceased to develop, except when they were cut just at the base of the forming disk. In such cases, however, new disks began to develop at once and sometimes two or three appeared upon the end of one mutilated stipe, Fig. 24.

To ascertain the effect of light or darkness upon the formation of the ascophore a flat was divided into four sections and planted December 14, 1906, with sclerotia. The first section was left uncovered, the second was covered with unprinted newspaper, the third with light weight manilla paper and the fourth with heavy weight manilla paper. Examination on February 5, 1907, showed that disks were fully formed upon section one, uncovered, though no disks showed in any of the other sections (Fig. 25). On February 27 the paper was all removed, and on March 11 very numerous disks were first observed on these newly uncovered sections. The time which elapsed between the removal of the paper and the appearance of the disks was considerably less than is usually taken to produce ascophores and it is

probable that the presence of the paper simply inhibited the formation of the disks, not of the stipe of the ascophore. Light is evidently the stimulus which causes the tip of the sprouts which come from

Fig. 24.—Ascophores, showing new disk formation after mutilation.

the sclerotium to stop growing in length and to expand into the disks and the presence of the paper covering was probably sufficient to exclude the light and thus to prevent this reaction.

As shown on page 118 heat or cold beyond certain limits retards or inhibits the formation of ascophores. The maximum temperature for ascophore formation seems to be about 8 to 25 degrees C.

The Ascospores.

The ascospores, as has frequently been noted, are discharged quite forcibly into the air when mature. To observe this phenomenon sclerotia-bearing disks were grown in flower pots and covered with plates of glass, or kept in tea cups covered with damp cloth. Upon the removal of the covering clouds of spores were forcibly ejected and could be followed by the eye for a distance of several feet. Currents of air caused by any one passing rapidly by ripe disks in open pots caused the ejection of spores. Spores for study were readily secured at any time by placing

a cover glass over a ripe disk, then breathing lightly upon it, resulting in an immediate and copious discharge of spores upon the cover glass.

To determine the per cent. of germination of ascospores and the toxic effect of copper sulphate, Bordeaux-mixture-filtrate and other substances hanging drop cultures were made in lettuce broth, in distilled water and tap water, also in various toxic substances. The tests were

FIG. 25.—Flat showing effects of different coverings upon the development of apothecia. I, Not covered; II, covered with heavy manila paper; III, covered with light manila paper; IV, covered with unprinted newspaper.

made by catching spores upon cover glasses by the method described above and preparing hanging drops with these, using the solution to be tested. The percentage of germination was read when the spores ceased to germinate.

Bordeaux filtrate No. 1 was made by merely filtering freshly prepared Bordeaux Mixture. The residue remaining upon the filter paper was then allowed to dry and remain thus one week. It was then washed with the least possible quantity of water and this wash water filtering through was tested as Bordeauz filtrate No. 2.

The result of these tests are shown in the following table:

TABLE X.—SHOWING EFFECT OF VARIOUS SUBSTANCES UPON SPORE GERMINATION.

Germination, Per Cent	Medium used.
68	Water distilled from Bichromate of Potash.
80	Distilled Water with Lampblack added, then filtered.
85	Ordinary Distilled Water.
92	Tap Water.
100	Lettuce Broth.
0	Copper Sulphate solution $\frac{n}{100}$
0	Copper Sulphate solution $\frac{n}{200}$
0	Copper sulphate solution $\frac{n}{800}$
10	Copper Sulphate solution $\frac{n}{1600}$
20	Copper Sulphate solution $\frac{n}{3200}$
50	Copper Sulphate solution $\frac{n}{6400}$
0	Bordeaux Mixture, Filtrate No. 1.
100	Bordeaux Mixture, Filtrate No. 2.

It is seen here that germination was more in ordinary distilled water than in the supposedly non-toxic water (U. S. Bureau of Soils) and that still higher germination was attained in tap water and higher yet in the nutrient lettuce broth. Copper sulphate N/800 is fatal to all of the spores and only 10 per cent. germination was found in the N/1600. The per cent. of germination increased almost in proportion to the weakening of the solution beyond this point. In the case of the Bordeaux filtrate it is seen that the first filtrate was fatal to all the spores while the second did not prevent germination at all.

To test the longevity of ascospores, collections were made on cover glasses and placed away in sterile petri dishes. These were tested in duplicate or triplicate the first and second day and every seventh day thereafter until no germination was apparent, using either lettuce broth or 4 per cent. sugar solution as the medium. The results of these tests appear below:

TABLE XI.—SHOWING LONGEVITY OF ASCOSPORES.

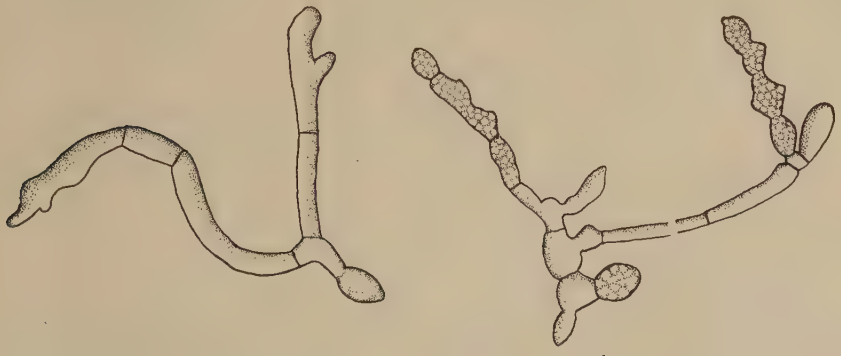
Age in Days.	1	2	7	14	21	28	35	42	49	56	63	70	77	84	91
Per Cent of Germination.	100	100	25	50	50	65	10	50	50	90	75	95	90	90	50
Medium Used in Cultures.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	4% Sugar.	4% Sugar.	4% Sugar.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	4% Sugar Solution.

TABLE XI—Continued.

Age in Days.	98	105	112	119	126	133	140	147	154	161	168	175	182	189
Per Cent of Germination.	90	75	75	50	75	50	25	10	1	0	0	0	0	0
Medium Used in Cultures.	4% Sugar Solution.	4% Sugar Solution.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.	Lettuce Broth.

While there is considerable fluctuation in the results the gradual falling off in germinations after the 125th day and the total lack of germination after 154 days or about 5 months seems to indicate that the ascospores do not live longer than this time. It is to be noted that the spores in these instances were kept dry. If they were allowed to become moist, as through absorption of water from a humid atmosphere, they immediately germinated. It is thus apparent that their only condition of longevity is in a dry state and that the results reported above therefore, represent their maximum life. Under other, perhaps under all normal conditions, their life is undoubtedly short.

In some cases cultures of ascopores in the sugar solution were examined after 26 hours. It was found that while at first growth had



a *b*

Fig. 26.—Germinating ascospores. *a*, Normal; *b*, abnormal from sugar solution.

been normal, Fig. 26a, very much distorted hyphæ, Fig. 26b, occurred in abundance and that the distorted cells were vacuolate after the manner of old hyphæ.

Infection by Ascospores.

The question of infection conditions is one of highest importance. Many tests were made to determine under what conditions ascospores could produce infection. The following trials are typical:

A number of fresh, healthy lettuce leaves was placed in culture dishes and inoculated with ascospores in the ways indicated below.

The checks were exact parallels of the inoculations but without spores. The results of these inoculations are shown in the following table:

TABLE XII.—REGARDING INFECTION BY ASCOSPORES.

Number of Leaves Inoculated	Methods of Inoculation	Surface of Leaf Inoculated	Number of Leaves Infected in 7 Days
10	Ascospores in lettuce broth	Upper	6
10	Ascospores in lettuce broth	Lower	0
10	Ascospores in water on bruised spot of leaf	Both	5
10	Check	Upper	0
10	Check	Lower	0
10	Ascospores in water	Upper	0
10	Ascospores in water	Lower	0
5	Ascospores at needle-prick	Upper	0
5	Ascospores at needle-prick	Lower	0

It is noted that the only infections occurred in the presence of the nutrient lettuce broth or at the bruised spots where, virtually, broth existed. This lack of ability of the spores alone, or in water merely, to infect was of such importance that repeated tests of this point were made.

Thus on November 21, 1907, ten lettuce leaves were inoculated upon the upper surface with ascospores in sterile water and on December 3, 1907, there was still no sign of infection.

On November 21, 1907, fifteen lettuce leaves were inoculated upon the lower surface with ascospores in sterile water and on December 3, 1907, there was still no sign of infection.

In both of these cases the leaves, kept in a damp chamber in ordinary room temperature, were fresh and healthy at the end of the experiment, even at the marked spots where the spores were placed. On December 4, 1907, spores were washed from the leaves which had been inoculated but which had shown no infection. Upon microscopic examination in all cases where the spores were found it was seen that they had germinated in a normal way but with no branching of the mycelium or flattening of its end. It was thus made clear that the lack of infection was not due to non-viability of the spores.

On December 4, 1907, sixteen lettuce leaves were inoculated in a damp chamber with spores placed in lettuce broth together with small pieces of lettuce leaf; of these, nine leaves became diseased in seven days while seven showed no infection. At the same time sixteen more leaves were prepared in the same way except that sterile water was used in place of the lettuce broth. In this case none of the leaves showed infection

on the seventh day and it was not until the twelfth day that any signs of infection were seen. The leaves had by that time wilted badly and these three cases can not be regarded as cases of parasitism.

To still further study infection by ascospores leaves were inoculated as is indicated in Table XIII. In the case of the inoculations with mycelium the vigorously growing mycelium was placed upon cover glasses in a drop of lettuce broth so that only the tips of the mycelium could touch the leaf by growing over the edge of the cover glass. The leaves were kept in culture dishes so as to preserve a humid atmosphere.

TABLE XIII.—REGARDING INFECTION BY ASCOSPORES.

Number of Leaves	Surface of Leaf	Ascospores + Sterile Water	Ascospores + Lettuce Broth	Mycelium
10	Upper	No leaves infected	No leaves infected	10 leaves infected
10	Lower	2 leaves infected	10 leaves infected	10 leaves infected
10	Bruise	2 leaves infected	10 leaves infected	10 leaves infected
10	Control	No leaves infected	No leaves infected	No leaves infected

Summarizing all of the experiments upon this point, it appears that direct infection by ascospores seldom, if ever, occurs but that infection from the mycelium follows in 100 per cent. of the cases. Spores placed upon a lettuce leaf in lettuce broth with a small bit of torn lettuce leaf also gave a reasonably high per cent. of infection. The use of a drop of lettuce broth upon the leaf in which to place the spores also usually gave infection. In only two cases, however, of all the trials made did any infection result from placing spores upon the leaves in pure water or upon the bare surface of the leaf. In view of this very small percentage of positive results we are not willing to accept this as evidence that the fungus can ever enter upon its parasitic existence without at first having attained vigorous headway saprophytically.

Parasitism and Saprophytism.

The fungus is clearly a saprophyte under many conditions as is attested by its luxuriant growth on various nutrient media and upon dead organic matter. That it may be parasitic as well is obvious from its inroads upon lettuce in our experiment beds and in many commercial beds of this State. The exact degree of its parasiticism and the extent to which it can exist as a saprophyte can, however, only be told by careful experimental work. Since these relations are of great economic significance, with strong bearing upon methods of prophylaxis, close attention was given to ascertain under what conditions and at what stages of its development this fungus is capable of paracitism and how long and upon what nutrients it can exist as a saprophyte, and in particular to what extent it can exist in and migrate through soil.

The following additional test of ascosporic infection was made. Thirty lettuce leaves were placed in culture dishes and upon the first

ten was placed sterile soil, upon the second ten sterile soil with ascospores upon the soil. At the edges of ten other leaves small pieces of manure were placed with ascospores on the edges of the manure which were farthest away from the lettuce leaf.

There was no infection in either the first or second case through soil while in the third case the spores germinated, grew over the manure and speedily caused the disease upon each of the ten leaves. It appears from this and other experiments previously quoted that ascospores are not capable of direct infection of the lettuce plant but

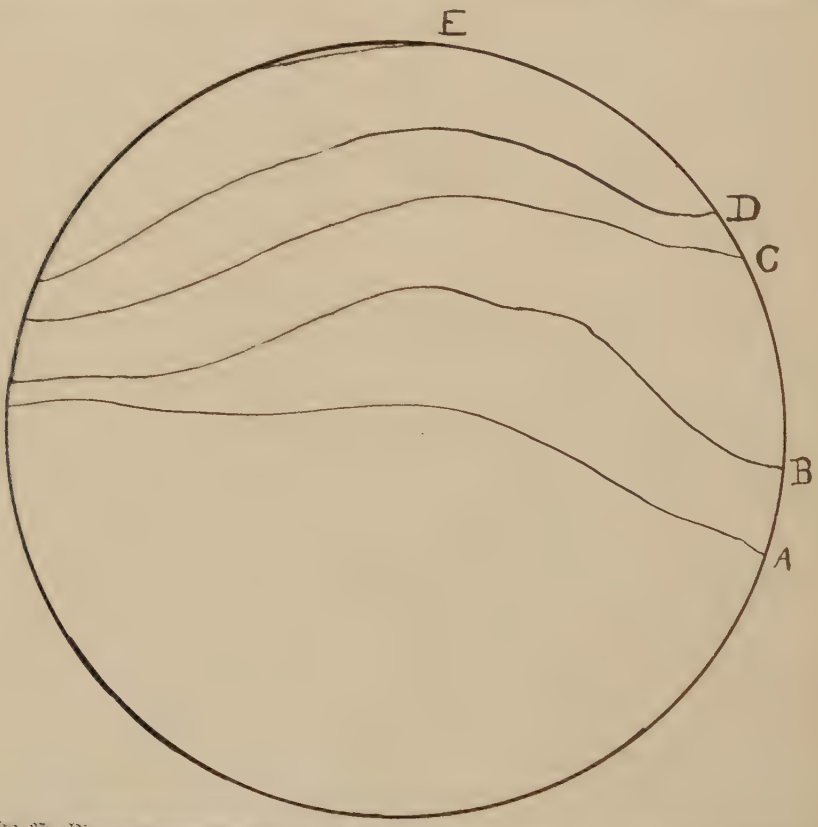


FIG. 27. -Diagram showing rate of invasion of a lettuce leaf by Sclerotinia. Each zone represents one day's growth.

that the germ tubes formed from the ascospores must acquire a considerable strength and develop a vigorous mycelium by a saprophytic habit before they can bring about infection of healthy tissue. Even the nutriment present in ordinary rich soil will not give the mycelium vigor sufficient to produce infection.

To fully demonstrate the parasitism of the ordinary vigorous mycelium the following test was made: On December 30, 1907, ninety-six plants in outdoor beds were inoculated at various places with masses of

vigorously growing mycelium, some placed as near the center of the head as was possible without tearing the plant seriously, others by laying the mycelium upon the lower leaves on the shaded side of the plant. All of these plants showed clear cases of infection at the end of fifteen days or earlier, and all the plants were dead and dried up on the first of April, 1908.

To determine the rapidity of growth of the mycelium of the fungus in a plant and its migration through the tissues of the plant a leaf, after it had become infected on one edge, was placed in a large petri dish and daily record was made of the growth of the mycelium by marking the outside of the plate. The daily growth as is shown by the accompanying diagram, Fig. 27, was on an average, about 11 mm., while the maximum for a single day was 15 mm. and the minimum for the same period 8 mm. The lines of the diagram indicating the growth each day, represent as nearly as possible, the place where the green and uninfected portion of the leaf joined the translucent or infected portion. The average temperature for the five days was 54.5 degrees F. with a maximum of 75 degrees and a minimum of 39 degrees.

After the mycelium exhausts the food supply from the portions of the plant which are attacked, it reaches into the air or over the earth; no aerial mycelium is noted upon newly infected tissue.

The Maximum Infection Distance for Mycelium.

A few lettuce leaves were placed on moist sand in a glass dish, inoculated and kept covered. After a vigorous growth had developed and the originally infected leaves were nearly consumed fresh leaves were placed at various distances from the infected mass. The maximum distance at which leaves were infected in this way, i. e., the maximum distance that the fungus could travel under such conditions, was 2.5 cm. White mycelium might be seen with the naked eye to a distance of about 8 mm. from its source of nourishment, no farther. A very few small sclerotia formed on the naked sand near the lettuce leaf.

In order to determine to how great a distance the mycelium would spread over strictly non-nutritious surfaces small pieces of lettuce leaf were laid in a large sterile petri dish and inoculated with mycelium. The dish was then placed in a moist chamber. The following is the record showing the distance to which the mycelium spread, using various amounts of lettuce leaf for inoculation.

It is evident that with a very small amount of lettuce from which to draw nourishment the mycelium spreads only a short distance. By increasing the amount of nutrient material the distances to which the mycelium can spread is increased up to a maximum of about 22 or 23 mm. It is thus apparent that the spread of this fungus over a soil to any great distance, unless the soil provide nutrient material, is not to be feared. It is noted also in this experiment with the small amount of nutrient material such as 50 sq. mm. of lettuce leaf no sclerotia are formed. With as much as 100 sq. mm., however, a sclerotium, though small, was made.

Mycelium was placed so that its tips only might come in contact with a lettuce leaf. In twenty-four hours it had grown to the leaf, and had developed holdfasts at each point of contact. Each holdfast was surrounded by a translucent spot 1 to 2 mm. in diameter. In twenty-four hours more the tissues were translucent to a distance of about 2 cm. in each direction from the point of original infection, though there was no development of mycelium external to the leaf except near the petiole where the nutriment afforded by the tissues was apparently exhausted.

TABLE XIV.—SHOWING DISTANCE OF MYCELIAL GROWTH, ON GLASS.

Amount of Lettuce Leaf	Distance of Mycelial Growth	Remarks
4 sq. mm.	4 mm.	About equally in all directions.
50 sq. mm.	7 mm.	
100 sq. mm.	8 mm.	Irregularly, one small pin-head-sized sclerotium was formed.
200 sq. mm.	18 mm.	Irregularly, one small pin-head-sized sclerotium was formed.
400 sq. mm.	23 mm.	Irregularly, one small pin-head-sized sclerotium was formed.
900 sq. mm.	22 mm.	Irregularly, one small pin-head-sized sclerotium was formed.

Numerous attempts were made to infect lettuce leaves with soil taken from beneath diseased plants and which was known to contain the mycelium, since it was clearly visible to the naked eye.

Thus, on February 8, 1908, fifty lots of such infected soil were taken was aseptic precautions by a sterilized spatula and placed upon healthy lettuce leaves in sterile culture dishes and dampened with sterile water. No infection resulted. It seems from these tests that the mycelium can not migrate far through soil and retain its infecting efficiency without additional organic food.

Hosts.

While this species of fungus has been reported upon many hosts it is notable that during all of our work of the past four years, involving the inspection of many infected beds in many localities, only two cases of the attack of this fungus in lettuce beds upon any plant other than lettuce have come under our observation. One of these cases was as follows:

A garden pea about 30 cm. high of vigorous though somewhat forced growth, was bent over into contact with the mycelium of the diseased lettuce leaf in the experiment mentioned on page 127. In eight days the distal end of the pea, some 12 cm. long, had been killed. Mycelium was breaking out through it in numerous wooly patches and sclerotia had begun to form. The fungus had also migrated some 9 cm. toward the root and for about 5 cm. the stem was covered with floccose mycelium (Fig 28). The invasion of the pea stem continued until the entire plant was killed.

In the other case B. B. Higgins observed a plant of Lamium which lay amid a mass of sclerotinized lettuce and had become infected. Both of these cases are exceptional. This rarity of infection of plants other than lettuce points somewhat strongly to specialization of the fungus or to very low resistance by the lettuce plant.

Sclerotinia libertiana has been reported on many other hosts and has been found in North Carolina upon a few others besides lettuce. B. B. Higgins brought it into the laboratory on cabbage and a sclerotium disease that appears to be identical with it was sent to the Ex-

FIG. 28.—Showing healthy leaves and two infected leaves lying upon soil; also an infected pea vine.

periment Station from Mebane by S. K. Scott upon crimson clover and by B. T. Pierce from Charlotte upon alfalfa. According to all appearances of the mycelial growth and formation of sclerotia these are identical but as the sclerotia were not large enough to produce asci and ascospores no comparative data on those structures were obtainable.

Among the hosts upon which the fungus has been reported are the following:

Upon Hemp in Russia, 1868.
" Potato in England, 1883.
" Bean in Germany, 1886, and in Holland.
" Petunia in Germany, 1886.

Upon Zinnia in Germany, 1886.
" Hemp in Alsace, 1891.
" Hollyhock in England, 1891.
" Cucumbers in Massachusetts, 1893.
" Mulberries in France, 1897.
" Sunflower, Dahlia, Eimia, Beets in Holland.
" Artichokes in France, 1899.
" Carrots in France, 1899.
" Chicory in France, 1899.
" Tomatoes in Massachusetts, 1900.
" Cauliflower in Missouri, 1905.
" Cabbage in Missouri, 1905.
" Artichokes in France, 1905.
" Forsythia in Switzerland, 1905.
" Lemon in California, 1907.
" Omphalodes in Switzerland, 1909.

Also upon Ægopodium, Cerefolium, beets, radish, chicory, mustard, rape, caraway, parsley, celery, fennel, vetch, clover.

Field Observations.

Study of diseased plants in New Bern in 1906 showed that the plants were generally attacked on the lower leaves at spots where they touched the soil. The fungus then worked both ways, toward the stem of the plant and toward the tip of the leaf, though it stops its migration in the latter direction very soon owing to the lack of moisture. Growing toward the stem of the plant it soon reaches the leaf axil and there forms masses of white cottony mycelium and eventually sclerotia. The greater number of sclerotia are formed here although some are formed at other points especially among the leaves that lie upon the ground.

Some time was spent sifting the soil, looking for sclerotia formed the previous year. From a bed that was badly infected it was difficult to get an average of more than one sclerotium from each two-quart sifter of soil.

Many germinated, disk-bearing sclerotia were found, nearly always in the front of the beds where the soil was shaded nearly all of the time. When disk-bearing sclerotia occurred in other parts of the beds it was always either in places where they could not be disturbed by cultivation or in beds that had not been recently cultivated. Germinated sclerotia were especially abundant under lettuce plants where they were both shaded and protected from mechanical disturbance. In the case of about half of the affected plants the diseased portion was either directly over a disk-bearing sclerotium or the faces of near by disks were turned toward the infected plant. In the latter case the plants invariably showed more disease upon the side of the plant nearest the disk. It was no uncommon thing to see clouds of spores discharged from disks in the field.

Disks were found upon sclerotia that were located at a depth of 1 1-2 inches in the soil. In no case were sclerotia found upon or

among the roots of a plant that had just died. The root was always still perfectly sound, even after the aerial portion of the plant was entirely dead and in no case were sclerotia found in the soil until the root had had sufficient time to rot.

General Relation of the Fungus in Nature.

From the facts above adduced experimentally and from field observation it is seen that *Sclerotinia libertiana,* which is the actual cause of the disease under discussion, propagates and spreads by means of its ascospores produced from the germinated sclerotia and by its mycelium.

The ascospores are comparatively short-lived, even under the conditions most favorable to their longevity. In condition of nature, subject to alternate dryness and atmospheric humidity they would invariably germinate and in the absence of favorable nutrient pabulum, die. Thus the ascospores can not function to any large degree as a means of carrying this fungus over periods of time of any considerable duration.

The mycelium has also been shown to be of comparatively short-life and to retain its infecting power but a short time in the absence of nutriment. This, too, can not be regarded as a means of carrying the fungus over long time intervals. Any possibility that the mycelium may remain alive in soil that has borne sclerotinized plants is very remote, probably non-existent. The sclerotium is long-lived and well adapted to perpetuate the fungus. It alone, of all the structures of the fungus, is able to live for sufficiently long periods to bridge over seasons adverse to the growth of the fungus or long periods of time when no food is available.

The fungus may, therefore, be likened to an annual plant, all parts of which except the seed die at the approach of winter, the sclerotium in this instance acting the rôle of the seed.

The sclerotia rest during the period of inactivity of the disease, that is from the harvest of one lettuce crop until the next crop is present under suitable weather conditions to permit of infection. They then germinate and produce apothecia with a crop of myriads of ascospores.

These ascospores have been shown unable to directly infect healthy lettuce leaves. They may germinate in the film of dew upon the plant but can not force entrance into its tissue and can only perish. Neither are mere wounds ordinarily sufficient to allow infection. The ascospores must at first sustain a period of saprophytic existence until the mycelium developing from it attains a certain degree of vigor. To do this the ascopores must fall upon and germinate upon organic matter, dead moist leaves or other plant parts or particles of manure or some other similar dead organic substance. A dead lettuce leaf or torn fragment of leaf serves its purpose admirably. If the saprophytic existence of the sporeling be upon organic matter which lies very near a lettuce plant the vigorous mycelium may reach over to it and parasitize it.

In cases where the ascospores fall upon dead lettuce leaf fragments still in contact with the live lettuce plant the infection bridge is open. If the ascospores fall upon organic matter separated by some centimeters from the lettuce plant the organic matter there available will be consumed and the mycelium will perish without any infection resulting. In some instances when the food supply is fairly large new sclerotia may be formed. Migration of the mycelium through the soil in efficient infecting condition, for any considerable distance, does not occur, even in soils bearing a large amount of organic matter as reckoned by the horticulturist.

Infection from plant to plant is governed by these same conditions. If the infected plant lie sufficiently near to the uninfected one, the distance can be bridged by the mycelium, but this does not occur at any great distance, usually not over a few centimeters.

That infection from plant to plant does not bridge over any large distance is shown by many observations of healthy plants standing, surrounded on all sides, by dead sclerotinized plants.

Attempts to Control the Disease.

According to Stone[9] the lettuce sclerotiniose became so bad in many green-houses in Massachusetts that many growers lost practically their whole crop from this cause. Disinfection of the soil, however, proved practicable under green-house conditions.

Two principal methods of soil disinfection were early used (1) by means of chemical solutions; (2) by means of heat. The first of these methods has been proved to be unsatisfactory and impractical in most cases. The second has been of a very great benefit.

One of the earlier methods for heating the soil was to sprinkle it with hot water. This was of some value but not entirely effective and moreover it possessed the disadvantage that it left the bed so wet that it had to remain idle for considerable time in order to become dry enough to be worked.

The substitution of steam as a disinfectant followed and it is today used with considerable success under green-house conditions. There are three ways in which steam may be used: (1) by placing 2-inch drain tile permanently under the soil in rows 16 inches apart, more or less as circumstances dictate, and flooding them with steam; (2) by laying perforated steam pipes upon the soil in the middle of the bed and throwing the soil from the sides upon them, then heating by steam, and after steaming pulling out the pipes to use elsewhere. The soil is then covered with canvas for several hours; (3) by a harrow-like arrangement of pipes. Fig. 29. The teeth of the harrow are perforated on all sides to allow the steam to escape into the soil. The teeth are driven into the soil to a depth of perhaps 10 inches and heated to 208 degrees F., and this temperature maintained as long as is desired.

The disinfection of green-house soil in some of these ways has been used in Rhode Island,[20] Vermont,[21] Ohio,[22] and some other States, nearly always with beneficial results.

In Kentucky,[11] a drop* is reported to have been checked by sub-irrigating and placing a mulch of excelsior under the plants to prevent their touching the soil.

FIG. 29.—Steaming the soil by means of Sargent's sterilizer.

To test the applicability of the above well-known methods to the cold frame and field conditions of the South, numerous experiments were planned and carried out at New Bern and West Raleigh, some of which were as follows:

Experiment 3. To test the efficiency of soil disinfection by heat. One-half of a bed designated as Bed A was prepared by laying 2-inch drain tile 10 inches deep and about 13 inches apart with the ends

FIG. 30.—Showing the arrangement of tile in box used for steaming the soil for top dressing. Ends of tile, *b*, were closed with cement.

all running into cross tile as shown in Fig. 30. Thermometers were placed in the soil, care being taken not to place any of them over the rows of tile.

*It is uncertain whether this was sclerotiniose or some other form of drop.

TABLE XV.—SHOWING TIME AND TEMPERATURE IN SOIL STERILIZATION EXPERIMENTS.

Time	12:30	12:45	1:00	1:15	1:30	1:45	2:00	2:15	2:30	2:45	3:00	3:15	3:30	3:45	4:00	4:15	4:30	4:45	5:00	5:15
Therm. a	8	10.5	14	21	25	35	45	46	55	65	74	78	85	90	95	60	70	89	95	95
" b	8	10.5	13	17	21	25	31	38	40	45	50	56	59	63	66	69	72	75	76	80
" c	9	9	10	14	20	30	40	57	70	80	89	94	51	63	85	90	95	95	95	95
" d	7	10.5	22	30	36	46	50	55	60	65	70	74	76	85	50	69	85	90	90	95
" e	8.5	9	9	9	11	11	13	15	15	17	22	28	33	41	57	80	92	95	95	95
" f	10	11	11	11	11	11.5	12	13	14	16	18	19	23	38	67	90	92	95	95	95
" g	7.5	8	8.5	8.5	9	9	9	10	10	10	11	11	12	14	20	28	60	74	80	90
" h	8	8.5	9	9	9	9	9.5	9.5	10	10.5	10.5	11	11.5	12	12.5	20	40	59	72	80
" i	8	8.5	8.5	9	9	10	10	10	10	10	10	10.5	11	12	21	60	95	95	95	95
Steam pressure in lbs.	65	60	60	60	58	45	60	60	60	60	65	60	60	60	65	60	60	60	60	60

Records were taken of the temperature at each thermometer every 15 minutes from the time steam was turned on until the end of the steaming, also of the steam pressure on the boiler (see table).

In this bed were planted 1,260 lettuce plants, care being taken to prevent the carrying of the disease into the bed from other infected beds; of these plants 88 or 6.98 per cent. had the characteristic disease. In the control bed, B, of 1,260 plants there were only 38 that showed the disease. This discrepancy may be accounted for by the fact that infection was worse in Bed A, before it was treated; but it is evident that such steaming did not give promise as a method of control.

Beds G and H were steamed with the Sargent Sterilizer, Fig. 29. The sterilizer was covered with heavy canvas during use. Beds G and H were each 30 feet long, and together they contained 696 plants. The soil was heated to 90 to 95 C., in less than 10 minutes, and that temperature maintained for thirty minutes. Of the 696 plants, 33 or 4.8 per cent. showed the disease, and again considerably more than in the control.

Experiment 4. To test the efficacy of top dressing with disinfected soil. The ends of three separate beds, C, B, and E, were used because these were more convenient to the steam connection with the boiler house.

A box was made 2 x 6 x 4 feet, thus capable of holding 48 cubic feet of soil. Soil was skimmed from the surface of the three beds; to a depth of one inch from E, of two inches from D, and of 3 inches from C, making in all a total of 150 cubic feet or approximately three times the box full. Soil was placed in the box to half fill it, then a system of drain tile was laid upon this soil and enough more soil was placed upon the tile to fill the box. Steam was turned on through the inlet, the ends of the drain tile being stopped with cement. The soil was heated to 90 degrees C. and kept at that temperature for one hour. This soil, after heating, was then scattered over the beds from which it had been taken and the disinfecting box filled again. Care was taken to sterilize the shovels each time after unsterilized soil had been handled with them.

The results of this top dressing of the beds is shown in the following table.

TABLE XVI.—SHOWING EFFECT OF TOP DRESSING WITH STEAMED SOIL

Bed	No. of Plants in Bed.	Healthy Plants	Diseased Plants	Percentage of Disease	Depth of Top Dressing
E	320	399	21	7.02	1 inch
D	320	304	16	5.26	2 inches
C	320	308	12	3.89	3 inches

It is seen that the disease decreased with the increase in depth of the disinfected top dressing.

Experiment 5. To determine the effect of carbon bisulphide, one litre per square metre. One-half of Bed F was treated with carbon

bisulphide, and covered with canvas for forty-eight hours, then the canvas was removed and the bed left open. No plants were set until the odor of the carbon bisulphide in the soil had disappeared.

At the time of harvesting the crop there had appeared in the bed 8 diseased plants in a possible 160, which is a slightly smaller per cent. than occurred upon the bed that was top dressed one inch deep. While this treatment seemed to check the disease somewhat it can not be regarded as satisfactory.

Experiment 10. To determine the effect of formalin 1 part to 400 parts of water.

Upon a bed fifteen long formalin of the above strength was sprinkled with a watering pot at the rate of one-half gallon to the square foot. One-half of this solution was applied at one time and the second half two hours later. After sprinkling the bed was left 2 days then stirred with a sterile hoe to aid in drying. Two days later the plants were set out. At harvest time the plants looked better, were more uniform in size, and there were no diseased plants among 160 that the bed contained. The treatment looked promising and the experiment was repeated at West Raleigh in 1908-09, with the result, however, that 62.5 per cent. of the plants were affected.

TABLE XVII.—SHOWING EFFECTS OF SOIL DISINFECTION.

Method	Number of Plants Treated	Diseased Plants in Treated Area	Healthy Plants in Treated Area.	Per Cent of Diseased Plants
Steamed with Drain Tile	1,260	88	1,172	6.98
Steamed with Sargent Sterilizer	696	33	663	4.80
Top Dressed with 1 in. Disinfected Soil	320	21	299	7.02
Top Dressed with 2 ins. Disinfected Soil	320	16	304	5.26
Top Dressed with 3 ins. Disinfected Soil	320	12	308	3.89
Sprinkled with Carbon Bisulphide	160	8	152	5.00
Sprinkled with Formalin at New Bern	160		160	
Sprinkled with Formalin at West Raleigh	144	90	54	62.50
Control	1,260	38	1,222	3.02

From the above experiments in attempts at soil disinfection little hope is to be had. Though these means of disinfection may apply well in green-house conditions, it is obvious that they are not satisfactory in extensive cold frames. Whether some other method of soil treatment may not be devised, we can not, of course, say. Theoretically, it seems possible, but that these means mentioned above are of practical service, seems very doubtful.

Practical Considerations.

The rational method of eradication of this pest based upon the facts here adduced would seem to be the same as that practiced against an

annual plant, which if not allowed to produce new seeds will eventually, upon the growth of all old seeds, be brought to the end of its existence.

Sclerotia are not formed in diseased plants until the nutriment in the part affected is consumed, that is not until the plant has given clear easily discerned evidence of the presence of the parasite. If all such plants be pulled and burned no sclerotia will be made. The few small roots remaining in the ground will not usually be affected at this stage and even if they are, the formation of sclerotia in the roots under the ground is not common. This practice of constant removal of the sclerotinized plants will prevent the formation of new sclerotia and in course of time the sclerotia already present in the soil will have either decayed or germinated and thus become harmless. As an additional precaution it is well to kill all mycelium which may be left in the locus of the removed plant by a liberal application of some disinfectant such as copper sulphate. It appears that this line of treatment is the most promising in cases where the destruction of the sclerotia by steam is not practicable.

Since the ascospores are harmless except through the threshold of dead organic matter upon which to begin growth as saprophytes the careful removal from the bed of all torn, injured, sick or dead lettuce leaves should be practiced and manure and organic matter of other kinds had best be removed from the surface of the beds in the neighborhood of plants or covered so as to remove them from ascosporic infection. Similarly any injury to the plants as tearing the leaves, etc., which would result in dead bits of leaf, should be scrupulously guarded against.

Since frequent stirring of the soil inhibits production of ascospores it will be well to rake over the top soil to a depth of a half-inch once each week.

The above methods are of general application either in field or greenhouse. In addition to this soil disinfection, though it has not proved practicable in field or under canvas, is of great value under glass.

Botrytis and Sclerotinia.

Some species of Sclerotinia are genetically connected with Botrytis as a conidial form, notably:

Sclerotinia fuckeliana has as conidial form *Botrytis cinerea* Pers.

Sclerotinia vaccinii has as conidial form a Monilia.

Sclerotinia cinerea Sch. has as conidial form *Monilia cinerea Bon.*

These facts together with the sometime association or botryose and sclerotiniose upon the same plant very naturally led to the assumption that the Botrytis of the lettuce sustained genetic connection with Sclerotinia libertiana upon the lettuce.

Conflicting views have been held upon this point. Thus DeBary[23] as early as 1886 held that *Sclerotinia limertiana* had no true conidial form while some other species of the genus did produce Botrytis conidia. Humphrey[5] of the Massachusetts Agricultural Experiment Station says that the rotting of lettuce is due to *Botrytis vulgaris* Fr. which "is with little doubt the conidial stage of some sclerotium pro-

ducing Peziza (Sclerotinia)." Stone and Smith[8] of the same station accepted this conclusion, but in 1898 and 1899 an extended study of *Sclerotinia libertiana* led them to believe that the Botrytis upon lettuce was not the conidial form of this Sclerotinia. Ramsey[13] of Wisconsin in 1904 considered the "drop" (Sclerotinia) as distinct from Botrytis. Hume[12] of Florida in 1901 believed this Sclerotinia to be entirely distinct from Botrytis.

Finally, Wulff,[24] also Westerdijk,[25] in recent papers upon these fungi, accept the conclusion that *Sclerotinio libertiana* has no Botrytis conidial stage.

Our experiments and observations bring out clearly the following facts:

1. That sclerotiniose may prevail in beds for months with its characteristic white mycelium with no accompaniment of botryose.
2. That botryose may similarly prevail in other beds with no accompaniments of sclerotiniose.
3. That in all of our cultures of these two fungi extending over an aggregate of some seven years, involving thousands of tube and plant inoculations, there has never occurred an instance of apparent change, in any way, from one of these forms to the other.
4. That the sclerotia of Sclerotinia differ in size and general appearance from those of Botrytis, and that in cases of botryose the sclerotia of Sclerotinia are not produced.
5. That the sclerotia of Botrytis invariably produce hyphæ and conidia upon germination, and that the sclerotia of Sclerotinia never do so.
6. That the sclerotia of Sclerotinia invariably produce ascospores, or at least abortive attempts to do so, while the sclerotia of Botrytis never do so.

We believe therefore that the evidence is sufficient to warrant the conclusion that these two fungi and the diseases caused by them are distinct and that one bears no present relation to the other, whatever their phylogenetic relation may be.

PART II.*

A Practical Test of a Curative Treatment.

The conclusions as recorded above, deduced from several years of laboratory and field study of this disease, especially those conclusions which point to the sclerotia as the only means of hibernation, began to force themselves upon the mind of the senior author some years ago. Those conclusions seemed to be so unavoidable and their logical effect upon horticultural practice so fundamental and so significant that it was deemed imperative to put the question to a crucial test. If the theory as enunciated be true, all that is necessary in order to rid a Sclerotinia-infected lettuce bed of its pest is to prevent the formation of new sclerotia in it for a period of two or perhaps three years.

To make such a test the first essential was a bed thoroughly and unquestionably infected and so located and managed that it would not be subject to aerial or other extraneous infection.

The experimental lettuce beds of the Experiment Station located on the farm at West Raleigh are reasonably well isolated from any other infected beds which might furnish air-borne ascospores to bring about reinfection. Precaution could easily be taken to prevent access of sclerotia through manure or other sources. The beds are two in number, each 208 feet by 9 1-2 feet in size, 30 inches high on the north side, 8 inches on the south side, and are covered in the usual way by canvas supplemented when need be by burlap mats. They accommodate eight rows of plants, 77 plants to the row, with a total capacity, therefore, of 1,232 plants. The beds were to some extent infected owing to the nature of the experimental work that had been conducted in them. This infection was not, however, considered sufficient to make the test crucial.

The first step, therefore, was to thoroughly infect the beds and to demonstrate that they were so infected and to secure a record of the degree of infection. This was accomplished in the spring of 1908 by inoculating several rows or about 67 plants of the then large nearly mature marketable lettuce with Sclerotinia mycelium. Within a few days, April 18, the plants so inoculated all collapsed and followed the usual course of the disease. These plants and considerable other lettuce refuse as well were allowed to remain on the ground and since the plants were large the number of sclerotia that remained on the soil was very great. Thorough infection seemed sure. The lettuce was followed by cucumbers and in the autumn of 1908, October, the crop was put in in the usual commercial way and the record of disease for that year presented in Table XVIII and in Diagram I (Fig. 31), shows clearly that a full and thorough infection had been produced.

The plants, it will be seen, began to die of sclerotiniose in December and in January they were dying rapidly. The last record of disease

*Printed in part in an earlier Bulletin. (26.)

was on March 16, 1909. A total of 545 plants had died of sclerotiniose or over 45 per cent. of those in the beds.

To make records of disease the beds were inspected carefully each day and the cause of disease was determined by culture or microscopic examination or both, so that no doubt could exist on this point.

The work so far demonstrates thorough infection of the beds.

The second step of the test consisted in removing the plants before new sclerotia could form in order to determine whether by so doing the bed could in the course of a few years be freed from infection.

TABLE XVIII.—DAILY RECORDS OF DEATHS FROM SCLEROTINIOSE, 1903-1909.

Date	Number of Plants Diseased	Date	Number of Plants Diseased
Dec. 3	1	Jan. 2	3
5	2	4	9
14	1	8	10
20	2	11	41
24	2	13	29
11	21	15	12
18	2	17	10
20	11	20	24
21	2	24	73
22	8	26	94
23	2	Feb. 1	10
25	5	5	33
26	8	8	6
2	4	10	57
28	3	12	41
29	4	16	25
		Total	555

The daily inspection was most rigid. All suspected case of sclerotiniose were closely watched and as soon as the symptoms became reasonably indicative of this disease the plants, entire, were removed to the laboratory. *Thus no sclerotia were allowed to mature in the beds.* As an additional precaution the locus of the diseased plants were sprayed with a strong Bordeaux mixture.

The lettuce was followed by cucumbers and in the autumn, October 15, 1909, the next crop of lettuce was set.

It now remained to see whether the disease had increased, as it would have done under the usual modes of handling, or whether a decrease had been brought about by the hygienic treatment that had been followed.

The record of disease was kept precisely as in the preceding year and the same methods were taken to prevent maturity of sclerotia.

The record is presented in Table XIX and in Diagram II (Fig. 31).

TABLE XIX.—DAILY RECORDS OF DEATHS FROM SCLEROTINIOSE, 1909-1910.

Date	Number of Plants Diseased
Feb. 18	1
March 4	1
22	1
28	1
31	2
Total	7

It will be noted that no plants died of sclerotiniose prior to February 18, 1910, and none after March 31, and *that in all only seven plants or one-half of one per cent. of the crop died.*

This very large decrease in disease under one year of hygienic treatment—about 99 per cent. of the disease had been removed—was more than was anticipated and indicates even shorter life of the Sclerotia and greater loss of sclerotia from rotting than was predicated for them.

The beds were again set with lettuce in December, 1910.

This year approximately the usual number of plants, or to be exact, 1,113 in all, were set. The crop was raised to maturity, cut and sold. The record of disease is shown in Table XXI and in Diagram III (Fig. 31).

TABLE XX.—DAILY RECORDS OF DEATHS FROM SCLEROTINIOSE, 1910-1911.

Date	Number of Plants Diseased
April 18	1
19	1
20	3
29	1
May 1	1
15	1

Whatever may be thought of the theoretical questions involved, certain practical conclusions stand forth clearly and unmistakably.

1. The lettuce beds were very thoroughly infected. See record of 1908-09, Table XVIII and Diagram I.

2. Under usual conditions and usual mode of handling this disease would not have decreased but would have increased or at least remained destructive during following years.

3. Under the treatment that was followed, which had been indicated as the proper one by our laboratory and field studies, the disease decreased very markedly after one year and remained unimportant during the second year.

Whether the treatment employed here can be expected always to give such satisfactory results under all conditions of soil and climate can not of course be stated.

DIAGRAM I.

WINTER, 1908-1909.

DIAGRAM II.

WINTER, 1909-1910.

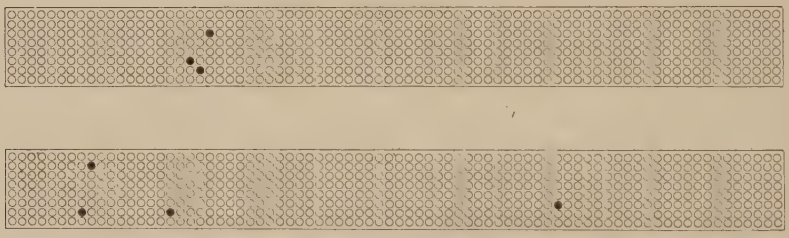


DIAGRAM III.

WINTER, 1910-1911.

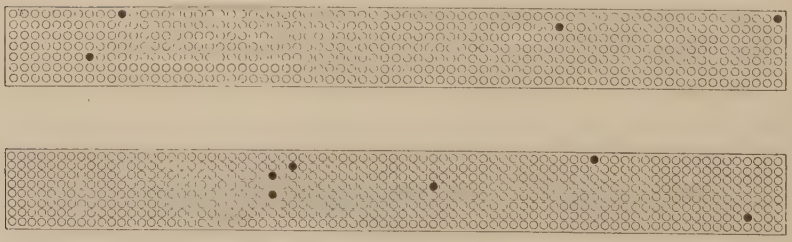


FIGURE 31.—Showing plan of Station beds and the exact location of affected plants in the bed during three seasons. • Diseased plant. o Healthy plant.

The writers feel justified, however, in stating that theoretically this treatment should prove effective and that in the one extremely crucial test to which it has been submitted it has proved thoroughly effective. They feel justified, therefore, in recommending it to lettuce growers who are troubled by this serious disease.

Recommendations.

The following recommendations for the treatment of beds infected with sclerotiniose are made.

1. The bed should be very carefully inspected every day and every plant that shows indications of this disease should be pulled up and burned.

2. The place in the bed from which sick plants are removed should be drenched with Bordeaux mixture or bluestone and water.

If these directions are followed no sclerotia will mature. The number of live sclerotia which will be present in the beds the following year will be very small and the amount of disease will be correspondingly reduced though it is not to be expected that the disease will be entirely eliminated. The next year the same treatment should be followed with just as much care as was given during the first year. Failure to be careful the second year will be fatal to success. It is probable that two years of this treatment will almost, if not quite, eradicate the disease. During later years, however, the beds should be watched closely and the same procedure followed. Beds which have been restored to a state of health and beds from which the disease has been partially eradicated should be protected from all possible sources of extraneous infection. It should be recognized that all refuse that comes from places where this disease exists is liable to bear the sclerotia and convey the disease. Therefore, all refuse from diseased lettuce beds, manure or fertilizer which may contain diseased refuse must be scrupulously avoided. There is also possibility of aerial infection. If infected beds exist nearby there appears to be no possibility of guarding against such infection and the method of treatment here advocated can not be expected to give its maximum of results if infected lettuce beds exist near the beds which are under treatment, since in such cases the danger of reinfection through the air will always be present.

BIBLIOGRAPHY

1. Kinney, L. F.: Garden Lettuce and Its Cultivation. Rhode Island Sta. Rpt. *10*: 270, 1898.

2. Philbroock, W. D.: Country Gentleman, *37*: 134, 1872.

3. Galloway, B. T.: Agricultural Science *8*: 304, 1894.

4. Smith, R. E.: Bot. Gaz. *29*: 369, 1900. Botrytis and Sclerotinia: Their Relation to Certain Plant Diseases and to Each Other.

5. Humphrey, J. E.: The Rotting of Lettuce. Massachusetts State Sta. Rpt. *9*: 219, 1892.

6. Bailey, L. H.: Lettuce. Cornell Univ. Exp. Sta. Bul. *96*: 387, 1895.

7. Selby, A. D.: Lettuce Rot. Ohio Sta. Bul. *73*: 221, 1896.

8. Stone, G. E., and Smith, R. E.: "Drop" of Lettuce. Massachusetts Hatch Sta. Rpt. *9*: 79, 1897.

9. Stone, G. E., and Smith, R. E.: The "Drop" of Lettuce. Massachusetts Hatch Expt. Sta. Rpt. *10*: 55, 1898.

10. Stone, G. E., and Smith, R. E.: Further Consideration in Regard to the "Drop" in Lettuce. Massachusetts Hatch Sta. Rpt. *11*: 149, 1899.

11. Garman, H.: A Method of Avoiding Lettuce Rot. Kentucky Exp. Sta. Bul. *81*: 3, 1899.

12. Hume, H. H. Florida Expt. Sta. Rpt. 1901: 91.

13. Ramsey, H. J. Wisconsin Sta. Rpt. *21*: 279, 1904.

14. Stevens, F. L.: Report of the Biologist. North Carolina Sta. Rpt. *30*: 16 and 66, 1907.

15. Stevens, F. L.: A Serious Lettuce Disease. North Carolina Sta. Press Bul. *14*.

16. Hutt, W. N.: Lettuce Growing in North Carolina. Dept. of Agr. Bul., Sept., 1907.

17. Stone, G. E., and Smith, R. E.: The Rotting of Greenhouse Lettuce. Massachusetts Sta. Bul. *69*: 1900.

18. Stevens, F. L., and Hall, J. G.: Variation of Fungi Due to Environment. Bot. Gaz. *XLVIII*: 1-30, 1909.

19. Saccardo, P.: Sylloge Fungorum *8*: 196.

20. Card, F. W., Blake, M. A., and Barnes, H. L. Rhode Island Sta. Rpt. 1906: 159-175. Plate 7.

21. Hills, J. H. Vermont Sta. Bul. *119*.

22. Selby, A. D. Ohio Sta. Circ. *57* and *59*.

23. DeBary. Bot. Zeit. *44*: 458, 1886.

24. Wulff, T.: Ark. Bot. *8*: No. 1-3, Art. 2, 18, Pls. 2, Figs. 4, 1909.

25. Westerdijk, J.: Med. u. h. Phytopath. lab. W. C. Scholton II Maart 1911.

26. Stevens, F. L.: N. C. Exp. Sta. Bull. *217*: 1911.